PHYSICIANS' GUIDEBOOK TO PUBLIC HEALTH LABORATORY SERVICES

1945

Connecticut State Department of Health
Stanley H. Osborn, M. D., C. P. H., Commissioner
Hartford, Connecticut

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PREFACE

For some time there has been a need for a publication that might be placed in the hands of every physician in Connecticut to furnish in "ready-reference" form information regarding the many diagnostic services that are now available at the Bureau of Laboratories of the Connecticut State Department of Health. This "Physicians' Guidebook to Public Health Laboratory Services" has been issued to meet that need. The volume is a contribution of the administrative staff of the Bureau of Laboratories. It has in no sense been hastily prepared but rests on a foundation of many years of laboratory experience buttressed by an appreciation of the many and intricate diagnostic problems that present themselves in the field of communicable diseases — a coign of vantage that can be attained only through frequent interchange of ideas with physicians, epidemiologists and health officers. The extent to which this little book is found useful will be the real test of its value to the physicians of the State.

The primary purpose of the undertaking has been to furnish information for each communicable disease under the headings of "Current laboratory services" that are available to physicians; "Collection of specimens" that are indicated worthwhile; and "Limitations of laboratory tests" that may be performed upon request. While we were engaged in the preparation of the manuscript, the Sixth Edition of "The Control of Communicable Diseases" was published by the American Public Health Association and it seemed to us that certain additional information from that volume would add greatly to the usefulness of this manual. Through permission kindly granted by Reginald M. Atwater, M. D., Executive Secretary of the American Public Health Association, it has been possible to include verbatim in the "Guidebook" the additional authoritative statements for each disease that appear under the headings*: "Etiologic agent", "Source of infection", "Mode of transmission", and "Prevalence".

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* "The Control of Communicable Diseases, An Official Report of the American Public Health Association" (6th Ed., 1945, published by the American Public Health Association, 1790 Broadway, New York, N. Y., (Price of single copies 35 cents) contains additional information for each of the communicable diseases of this comprehensive list under the headings: "Recognition of the disease", "Incubation period", "Period of communicability", "Susceptibility and immunity" and "Methods of control". That publication is suggested as a worthwhile companion to this "Guidebook" on the desk of every physician in Connecticut.

PREFACE

The puzzling question of which diseases to include in this compendium and which to omit was solved through a decision to include all of the diseases listed under the 72 sections in the "Control of Communicable Diseases". The reason for doing this is the seemingly sound one that the selection for that volume has resulted from a 20-year period of study by a carefully selected continuing committee of prominent authorities in the administrative control of communicable diseases. With the adoption of the list there are included in this volume some diseases that are rarely if ever seen in Connecticut; however, it would be presumptuous to say with respect to almost any disease included that it will not appear in our midst under post war conditions, at least as an isolated case. Under these circumstances we have preferred to err by including a disease that may never appear in this State rather than to omit data that may be of real worth to a physician in an emergency when the unexpected occurs. In addition, ten groups of diseases* have been added which appeared of importance from the standpoint of this publication.

The Connecticut State Department of Health offers this manual to the physicians of the State in the belief that it is as complete as is desirable to answer the questions that will ordinarily arise in regard to the laboratory tests currently available for public health purposes; as concise as such a work can be; as authoritative and up-to-date as any reference book available anywhere for the purpose. Comment and criticism will be welcomed. It is hoped that physicians generally will find the "Guidebook" useful enough so that instead of becoming a dust covered volume that is seldom or never used it will be a frequently consulted reference on the desk of many a physician.

FRIEND LEE MICKLE, SC. D.

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* The ten sections on "Cestode Infections, miscellaneous", "Gas Gangrene", "Haverhill Fever", "Hydatid Disease", "Mycotic Infections, miscellaneous", "Pinworm Infection", "Protozoan Diseases, miscellaneous", "Trematode Infections", "Trichuris Infection", and "Vincent's Infection (Trench mouth), Fusospirochetosis" appear only in the "Guidebook" and were not taken from the publication cited.

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OUTFITS FOR COLLECTION OF SPECIMENS

Specimen outfits are referred to in text by symbols. These outfits may be obtained from the Bureau of Laboratories upon request. When ordering, use symbols.

ACTINOMYCOSIS

1. *Etiologic agent.* *Actinomyces hominis* and other species of this genus.

2. *Source of infection.* *Unknown.* Possibly in some cases of actinomycosis in man, *Actinomyces hominis* previously existed as a saprophyte in the oral cavity (carious teeth, interstices between teeth, and crypts of tonsils).

3. *Mode of transmission.* Among cattle, principally by grains, grasses, and other cattle fodder, and stable bedding contaminated by discharges from lesions of the disease, infecting abrasions or wounds of oral cavity or body surface. It is not probable that the disease is transmitted from man to man. It may be transmitted from animal to man, but only rarely and indirectly through infection of oral or skin wounds by contaminated materials. The disease sometimes follows extraction of carious or broken teeth, or accidental injury, particularly to the jaws.

4. *Prevalence.* Infrequent among humans.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Microscopic examination of direct and stained smears from abscesses or other lesions for typical granules and for *Actinomyces.*

   B. Cultures of material from abscesses or other lesions for *Actinomyces.*

6. *Collection of specimens.* Collect pus from abscess in sterile bottle in "MI" outfit; if lesion is small, collect pus on sterile swab in "HS" outfit and plunge swab into agar jelly in tube furnished before submitting to laboratory. Also make smeared preparations on microscope slides using sterile swab ("VI" or "GC" outfit may be used for smears). In all cases, mark history blank plainly "For actinomycosis".

7. *Limitations of laboratory tests.* Presumptive evidence of infection by microscopic examination should be confirmed by culture whenever possible.

ANTHRAX

1. *Etiologic Agent.* Anthrax bacillus, *Bacillus anthracis.*

2. *Source of infection.* Hair, hides, flesh, and feces of infected animals.

3. *Mode of transmission.* Inoculation as by accidental wound or scratch, inhalation of spores, ingestion of insufficiently cooked meat, and mechanically by flies.

4. *Prevalence.* Rare and sporadic in humans and associated only with the occurrence of the disease in cattle, or with handling hide and hair products from infected animals. In epidemic form in cattle in various countries, such as Argentina and Manchuria, from time to time.

5. *Current laboratory services. Available at Bureau of Laboratories:* Isolation and identification of *Bacillus anthracis* by culture and animal inoculation of body fluids and of suspected sources of infection derived from animals.

6. *Collection of specimens.* Collect pus or other body fluid on sterile swab contained in "HS" outfit. Follow directions on history blank in "HS" outfit but mark history blank plainly "For anthrax". In suspected pneumonic infections submit sputum in sterile bottle in "MI" outfit.

   Before submitting specimens from suspected sources of infection contact Bureau of Laboratories.

7. *Limitations of laboratory tests.* Suspicious cultural findings must be confirmed by animal inoculation tests. This procedure is necessary to distinguish between *Bacillus anthracis* and common saprophytic spore-forming organisms.

   Since anthrax bacilli are not present in the blood stream in large numbers until just before death, a blood specimen is of value only during the last stages of the disease.

ASCARIASIS

1. *Etiologic agent.* *Ascaris lumbricoides,* the large intestinal round worm of man.

2. *Source of infection.* Excreta of infected persons, particularly children, and articles soiled with such excreta in and about houses lacking facilities for sanitary disposal of human wastes.

3. *Mode of transmission.* By direct or indirect transmission of the embryonated eggs from soil or other polluted material to the mouth. The embryonated eggs hatch in the intestinal canal, penetrate the wall, and reach the lungs by the circulatory system. Most of those which reach the lungs in the blood stream pass into the air passages, throat, and stomach, and thence to the small intestines. Polluted soil may be carried on the feet or footwear into houses and conveyances and to some distances.

4. *Prevalence.* High incidence of infection is found where low standards of hygiene, lack of sanitary essentials, poverty, and ignorance create the conditions conducive to intensive pollution of soil in the immediate vicinity of houses. Children of the runabout and early school age are likely to be more frequently and more heavily infected than are older children and adults. Particularly prevalent in the United States among the people of the Appalachian plateau.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic examination of feces for ova of *Ascaris lumbricoides* or identification of adult worm when passed.

6. *Collection of specimens.* With a tongue depressor, spoon, or other instrument collect specimen of feces, preferably from different parts of the stool, and place in bottle contained in "PD" outfit.

7. *Limitations of laboratory tests.* A series of stool specimens should be submitted for examination for intestinal parasites before placing too great reliance upon negative findings.

BARTONELLOSIS (OROYA FEVER, VERRUGA PERUANA)

1. *Etiologic agent.* *Bartonella bacilliformis.*
2. *Source of infection.* The blood of an infected individual.
3. *Mode of transmission.* Sand flies of the genus *Phlebotomus.*
4. *Prevalence.* This disease is limited largely to narrow mountain valleys of Peru. Recently has been discovered in Columbia and Ecuador.
5. *Current laboratory services. Available at Bureau of Laboratories:* Examination of blood smears for organisms morphologically typical of *Bartonella bacilliformis.*
6. *Collection of specimens.* Make thin blood films as for differential white cell count using slides in "MA" outfit. Allow to dry without application of heat. Mark history blank plainly "For *Bartonella*".
7. *Limitations of laboratory tests.* The findings of morphologically typical organisms in blood films is considered confirmatory of clinical evidence of this infection. Specimens taken at the height of the fever are the most satisfactory.

CESTODE INFECTIONS, MISCELLANEOUS (TAPEWORMS)

1. *Etiologic agent.* Among others are *Taenia saginata* (beef tapeworm), *Taenia solium* (pork tapeworm), *Hymenolepis nana* (dwarf tapeworm), *Dipylidium caninum, Diphyllobothrium latum* (broad Russian tapeworm).

2. *Source of infection.* Infected humans or animals which serve as primary host.

3. *Mode of transmission.* An intermediate host is required except for *Hymenolepis* where transmission may be direct. Infection occurs by ingestion of the eggs (direct) or of cysticerci in incompletely cooked meat or otherwise.

4. *Prevalence.* Widespread and common in the United States.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic examinations of feces for ova, and of proglottids when found.

6. *Collection of specimens.* Use "PD" outfit. Place portions of stool (proglottids when present) in bottle provided.

7. *Limitations of laboratory tests.* Identification of species from the ova is possible except for *Taenia solium* and *Taenia saginata* which cannot be distinguished from each other except by study of proglottids.

CHANCROID (SOFT CHANCRE)

1. *Etiologic agent.* Ducrey bacillus, *Hemophilus ducreyi.*

2. *Source of infection.* Discharges from lesions.

3. *Mode of transmission.* Chancroid is predominantly venereal in origin but has occurred rarely on the hands of doctors and nurses through professional contact with infected persons. It may similarly occur in children through accidental inoculation. Acquisition through indirect contact with articles soiled with moist discharges from the lesions of infected persons is rare.

4. *Prevalence.* Widespread but particularly common in subtropical and tropical areas among populations sexually promiscuous and living at a low economic and social level.

5. *Current laboratory services. Available at Bureau of Laboratories:* Examination of smears of fluid or exudate from lesions, preferably before they ulcerate. Dark field examination to exclude syphilitic infection is often advisable (See under "SYPHILIS").

6. *Collection of specimens.* Use "GC" or "VI" outfit. With sterile swab furnished collect material from lesion and smear on microscope slides. Mark history blank plainly "For chancroid".

7. *Limitations of laboratory tests.* The finding of typical organisms is considered confirmatory of clinical evidence of this infection.

CHICKENPOX (VARICELLA)

1. *Etiologic agent.* A specific filterable virus.

2. *Source of infection.* The infectious agent is presumably present in the lesions of the skin and of the respiratory tract; lesions of the latter, appearing early and sometimes inapparent, may render the disease communicable before the exanthem is in evidence.

3. *Mode of transmission.* Directly from person to person; indirectly through articles freshly soiled by discharges from an infected person.

4. *Prevalence.* Universal. Probably 70 per cent of persons have had the disease by the time they are 15 years of age. Not uncommon in early infancy. Winter and spring are seasons of greatest prevalence in North America.

5. *Current laboratory services.* No practicable laboratory aids to diagnosis known to Bureau of Laboratories.

CHOLERA

1. *Etiologic agent.* Cholera vibrio, *Vibrio comma.*

2. *Source of infection.* Bowel discharges and vomitus of infected persons, and feces of convalescent or healthy carriers. Contacts may become temporary carriers.

3. *Mode of transmission.* By water and raw foods; by contact with infected persons, including carriers, or articles freshly soiled by their discharges; by flies.

4. *Prevalence.* Absent in the Western Hemisphere (except when introduced from abroad). Appears occasionally in epidemic form in the Philippines. Prevalent in India and the Orient.

5. *Current laboratory services. Available at Bureau of Laboratories:* Cultural examination of feces for *Vibrio comma.*

6. *Collection of specimens.* Collect one or two ml. of the feces which are usually liquid ("rice-water stools") and place in the sterile bottle containing suitable preservative in the "FE" outfit. Mark history blank plainly "For cholera".

7. *Limitations of laboratory tests.* A few other organisms that closely resemble *Vibrio comma* may be differentiated from it by serological means.

CHORIOMENINGITIS

1. *Etiologic agent.* A specific filterable virus.

2. *Source of infection.* No evidence of person to person transmission. Reservoir of virus found in house mice *(Mus musculus musculus).*

3. *Mode of transmission.* The virus escapes from infected animals in mouth and nasal secretions, urine, and feces. Transmission to man is probably through infected food or dust, possibly occasionally by insects. Dogs, guinea pigs, white mice, and monkeys are susceptible to the virus.

4. *Prevalence.* Rare but more common than the number of recognized cases indicates.

5. *Current laboratory services.* Physicians may find the services of the Bureau of Laboratories helpful in eliminating meningitis of bacterial origin but no specific tests for choriomeningitis are available at present in the Bureau of Laboratories. In certain virus laboratories throughout the country this virus can be isolated from spinal fluid and neutralization tests made on blood from patients during illness and convalescence.

6. *Collection of specimens.* Contact Bureau of Laboratories before sending any specimen if virus studies are desired. Spinal fluid for bacterial meningitis may be submitted in "SF" outfit.

7. *Limitations of laboratory tests.* At present the lack of facilities for specific diagnostic aids constitutes the main limitation. Cell counts (preferably made locally) and total protein tests on spinal fluid may be helpful but are not specific.

   Isolation of the virus provides confirmatory evidence of infection. Increase in neutralizing bodies in the blood during convalescence as compared with the blood titer during illness is strong presumptive evidence of infection.

COCCIDIOIDOMYCOSIS (COCCIDIOIDAL GRANULOMA, "VALLEY FEVER")

1. *Etiologic agent. Coccidioides immitis.*

2. *Source of infection.* Dust, soil, and vegetation contaminated with the spores of the fungus.

3. *Mode of transmission.* Inhalation of spores in dust and dry vegetation, and, in laboratories, inhalation of spores from cultures. Infection through skin wounds is a possible but unlikely route.

4. *Prevalence.* "Valley fever" is prevalent in endemic areas in the southwestern United States and in the Chaco region of Argentina and Uruguay. Incidence highest in hot dry weather, most common in white females. Recovery is usually complete. Coccidioidal granuloma is of sporadic occurrence in endemic areas, most common in males. Case fatality about 50 per cent in the granulomatous form; only very few "Valley fever" cases progress to this form. The disease is to be considered in those who have been in an endemic area as well as in residents of such areas.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Microscopic and cultural examination of material from lesions.

   B. Microscopic and cultural examination of sputum in the pulmonary type of infection.

6. *Collection of specimens.*

   A. Collect pus in "MI" outfit for cultural and microscopic examination. (Use "TC" or "PN" outfit if "MI" is not available but, if so, plainly mark blank "For coccidioidomycosis".)

   B. Collect sputum that has been coughed up from the lungs. Place sputum in sterile bottle, preferably in "MI" outfit (If "TC" or "PN" outfit is used, history blank must be plainly marked "For coccidioidomycosis".).

7. *Limitations of laboratory tests.* Suggestive microscopic findings should be confirmed by cultural identification of the organism, *Coccidioides immitis*, and by animal inoculation.

COMMON COLD

1. *Etiologic agent.* One or more filterable viruses.

2. *Source of infection.* Discharges from nose and mouth of infected persons.

3. *Mode of transmission.* Usually directly by coughing, sneezing, and explosive manner of speech by which droplets are cast out into the air from the infected person to susceptible persons especially within short range; and indirectly by handkerchiefs, eating utensils, or other articles freshly soiled by discharges of the infected person.

4. *Prevalence.* Most persons, except those living in small isolated communities, have one or more colds each year. The incidence does not vary materially according to age, sex, race, or occupation, but incidence appears to be highest in children under 5 years of age.

5. *Current laboratory services.* No specific diagnostic laboratory aid available.

CONJUNCTIVITIS, ACUTE INFECTIOUS (OF THE NEW BORN, NOT INCLUDING TRACHOMA)

1. *Etiologic agents.* The gonococcus or some member of a group of pathogenic organisms, including the hemophilic bacilli and a filterable virus (inclusion blenorrhea).

2. *Source of infection.* Discharges from conjunctivae, or adnexa, or genital mucous membranes of infected persons.

3. *Mode of transmission.* Contact with an infected person or with articles freshly soiled with discharges of such person.

4. *Prevalence.* Occurrence varies widely according to the observance or neglect of prophylactic use of a solution of silver nitrate or equivalent preparation in the eyes of the new born by the attendant at the delivery. An infrequent complication in the present-day care of the new born.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic and cultural examinations of pus or exudate for identification of organism, especially for presence or absence of gonococcal infection.

6. *Collection of specimens.* Submit smear of pus or exudate on slides in "GC" outfit. When, in addition, gonococcal culture is desired, use "NC" outfit. See directions under "GONORRHEA".

   Material for general culture may be submitted on sterile swab in "HS" outfit; mark history blank "For identification of organism".

7. *Limitations of laboratory tests.* The finding of gram-negative intracellular diplococci is strong presumptive evidence of gonococcal infection subject to confirmation by culture. Isolation of common pyogenic bacteria may result from skin contamination. Certain forms of this disease are apparently of virus etiology (inclusion blenorrhea).

DENGUE

1. *Etiologic agent.* A filterable virus.

2. *Source of infection.* The blood of infected persons during first 5, usually during first 3, days of the disease.

3. *Mode of transmission.* By the bite of infected mosquitoes, *Aedes aegypti* (or *Aedes albopictus* in the oriental tropics), from 11 days after biting a patient until the death of the mosquito.

4. *Prevalence.* May occur wherever the vector *Aedes* mosquitoes exist, mainly in tropics and subtropics.

5. *Current laboratory services.* No diagnostic laboratory aid available at the Bureau of Laboratories.

6. *Collection of specimens.* Contact Bureau of Laboratories before submitting any specimen. Providing arrangements can be made for submission to a laboratory where immunological studies are being carried on for this disease, fresh specimens of blood should be drawn aseptically without preservative or anticoagulant, (1) during the acute stage and (2) during the convalescent state.

7. *Limitations of laboratory tests.* At present the lack of facilities for specific diagnostic aids constitutes the main limitation. A rise in the neutralizing power of the patient's serum against the virus during convalescence is strong presumptive evidence of the disease.

DIARRHEA OF THE NEW BORN, EPIDEMIC

1. *Etiologic agent.* Unknown.
2. *Source of infection.* Unknown.
3. *Mode of transmission.* Unknown, presumably direct or indirect person to person infection.
4. *Prevalence.* The disease is met with frequently in the United States and Great Britain, and is probably more widely spread. Epidemics occur slightly more frequently in the spring and summer months and always in nurseries for the new born.
5. *Current laboratory services.* No specific diagnostic laboratory aids available at Bureau of Laboratories but cultural examination of feces specimens for the exclusion of known pathogens is recommended.
6. *Collection of specimens.* For purposes suggested above under *"Current laboratory services"* submit specimens of feces in "FE" outfit. Using the sterile swab in the outfit, transfer a portion of the stool about the size of a sphere 1/2" in diameter to the specimen jar and emulsify it in the glycerol solution. If stools are liquid, transfer to the specimen jar with a clean spoon or other implement a quantity approximately 2 ml. in volume. Do not use spoon or other implement for more than one sampling unless carefully sterilized in the interim.
7. *Limitations of laboratory tests.* For the present, laboratory tests must be confined to those for the exclusion of known bacterial pathogens.

DIPHTHERIA

1. *Etiologic agent.* Diphtheria bacillus, *Corynebacterium diphtheriae* (the Klebs-Loeffler bacillus).

2. *Source of infection.* Discharges from diphtheritic lesions of nose, throat, conjunctiva, vagina, and wound surfaces. Secretions from the nose and throat of carriers of the bacillus.

3. *Mode of transmission.* Directly by personal contact, indirectly by articles freshly soiled with discharges, or through contaminated milk or milk products.

4. *Prevalence.* Endemic and epidemic. Two-thirds or more of the urban cases are in children under 10 years of age and two-thirds or more of the urban deaths occur in children under 5 years of age. More common in temperate zones than elsewhere, and in fall and winter months.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Microscopic examinations of throat and nose cultures for *Corynebacterium diphtheriae.*

   B. Determination of virulence (toxigenicity) of *Corynebacterium diphtheriae* by animal inoculation tests.

6. *Collection of specimens.*

   A. For microscopic examination of cultures: Use "KL" outfit. Rub the sterile swab marked "Throat" gently but firmly against any visible "false membrane". Then rotate swab gently over the coagulated blood serum medium in the vial marked "Throat" but do not break the surface of the medium. Do not discard the swab but replace it in the envelope and return it in the outfit with the culture. When no such membrane is present, rub the swab over the mucosa of the lower pharynx and tonsils before inoculating throat culture tube. Bacterial contamination may result if small food particles are picked up by the swab. Under no conditions

should any attempt be made to collect material shortly after application of antiseptics or germicides to the throat.

Then rub the sterile swab marked "Nose" gently over the mucous membrane of the nasal cavities. Cultures from the nostrils are more successful if the nostrils are first cleansed with physiological salt solution. Inoculate culture tube marked "Nose" and return swab as directed above.

B. Upon request, virulence tests will be performed on organisms isolated from specimens submitted for release or for detection of carriers of diphtheria. Mark history blank plainly "For virulence" when virulence test is requested.

7. *Limitations of laboratory tests.*

A. A positive report on a culture means that organisms morphologically typical of the diphtheria bacillus were observed. This is considered confirmatory of clinical evidence of the disease.

A single negative culture should not be considered conclusive if there are clinical symptoms. In all such cases another culture from the throat and also one from the nose should be sent. Do not delay administration of antitoxin if laboratory tests fail to confirm a diagnosis made upon clinical grounds. Negative reports on at least two successive cultures taken at least 24 hours apart from nose and throat are required for releasing contacts and cases of diphtheria.

B. A positive result of a virulence test means that the Bureau of Laboratories has determined that the culture produced gross pathology in the guinea pig typical of that produced by diphtheria toxin and that an animal protected by antitoxin was not similarly affected.

DYSENTERY, AMEBIC (AMEBIASIS)

1. *Etiologic agent. Endamoeba histolytica.*
2. *Source of infection.* The bowel discharges of infected persons.
3. *Mode of transmission.* By eating contaminated foods, especially those that are commonly served cold and moist, and hand-to-mouth transfer of the infected material from moist objects soiled with discharges of an infected individual; by drinking contaminated water; and by flies.
4. *Prevalence.* Not a common disease clinically recognized in continental North America. Epidemic outbreaks are rare. It is estimated that almost 5 per cent of the population are carriers of cysts. More prevalent in the tropics and the Orient.
5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic examination for trophozoites and cysts of *Endameba histolytica.*
6. *Collection of specimens.*

   A. In suspected chronic infections take portions of feces from different parts of the stool and place in bottle in "PD" outfit.

   B. In acute amebic dysentery, proceed as follows: Telephone Bureau of Laboratories that examination for trophozoites is desired so that preparations may be made for immediate examination of the specimen. The specimen must be delivered to the Bureau of Laboratories immediately after defecation (within 1 or 2 hours) and must not be chilled. A liquid stool passed after a saline cathartic is recommended.

7. *Limitations of laboratory tests.* Examination of the stool for vegetative forms (trophozoites) of amebae can be made only on fresh specimens submitted as described under *"Collection of specimens"*. A search for amebic cysts and for other parasites and ova can be made on older specimens. *Endameba histolytica* is generally regarded as the only pathogenic ameba and is the cause of amebic dysentery and its sequelae. *Endameba coli, Endolimax nana* and species of *Iodameba* are considered to be non-pathogenic.

   A single negative result is of little value for the exclusion of amebiasis; a series of stool specimens should be submitted.

DYSENTERY, BACILLARY (SHIGELLOSIS)

1. *Etiologic agent.* Various species of the genus *Shigella*, such as Flexner, Sonne, Shiga and others.

2. *Source of infection.* The bowel discharges of infected persons and carriers. Healthy carriers are common.

3. *Mode of transmission.* By eating contaminated foods, or drinking contaminated water and by hand-to-mouth transfer of contaminated material: by flies; from objects soiled with discharges of an infected individual or of a carrier.

4. *Prevalence.* Endemic, epidemic, and sporadic, but shares with other enteric infections the characteristics of a striking and progressive reduction wherever water supplies are rendered safe, sewage is disposed of in a sanitary manner, milk is pasteurized, and infant hygiene is of a good order. Most common in the summer months. Institutional outbreaks are frequent.

5. *Current laboratory services. Available at Bureau of Laboratories:* Cultural examinations of feces for the various types of dysentery bacilli. Facilities are also available for serological typing of cultures isolated at other laboratories which lack typing facilities.

6. *Collection of specimens.* Use "FE" outfit. Using the sterile swab in the outfit, transfer a portion of the stool about the size of a sphere 1/2" in diameter to the specimen jar and emulsify it in the glycerol solution. If stools are liquid, transfer to the specimen jar with a clean spoon or other implement a quantity approximately 2 ml. in volume. Do not use spoon or other implement for more than one sampling unless carefully sterilized in the interim. Care should be taken that the stool is passed into a vessel that has been thoroughly disinfected or sterilized.

7. *Limitations of laboratory tests.* Final identification of dysentery bacilli depends upon adequate serological tests. Unless such tests are performed, dysentery-like organisms may be mistaken for those which cause bacillary dysentery. The type reported is of importance in establishing relationship to other cases or to carriers of dysentery organisms.

   The examination of blood specimens for agglutinins is not recommended because many persons, particularly adults, may show confusingly high titres without evidence of infection.

ENCEPHALITIS, INFECTIOUS

1. *Etiologic agent.* Probably a virus for the Vienna type; a specific filterable virus for each of the other types.

2. *Source of infection.* Unknown. Birds are a probable reservoir of infection for the equine types.

3. *Mode of transmission.* In the case of the equine and the St. Louis types of encephalitis several species of mosquitoes have been shown to be capable of transmitting one or more of the viruses and mosquitoes are probably the important natural vectors.

4. *Prevalence.* The Vienna type was first distinctly recognized in 1917, but had occurred before, and has since been prevalent in many parts of the world, especially from 1920 to 1936, infrequently now. The St. Louis type was especially prevalent in the St. Louis area in 1933, where there was an incidence of 100 per 100,000 population but this type has occurred elsewhere before and since. The Vienna type occurs at all seasons of the year but more frequently in late winter and spring. The other types occur notably in late summer and fall.

5. *Current laboratory services. Available at Bureau of Laboratories:* Examination of spinal fluid for the exclusion of bacterial infections.

6. *Collection of specimens.* Contact Bureau of Laboratories before submitting any specimens. Instructions will be given if arrangements can be made for examination of specimens at a laboratory where virus studies are being made.

7. *Limitations of laboratory tests.* At present the lack of facilities for specific diagnostic aids constitutes the main limitation. Demonstration of neutralizing antibodies for a specific virus in the blood of an individual during convalescence or after recovery is presumptive evidence of infection.

   In fatal infections pathological changes in the brain tissue may be of value in determining type of illness.

FAVUS

1. *Etiologic agent.* *Trichophyton schoenleini (Achorion schoenleini).*

2. *Source of infection.* Lesions of skin, particularly of scalp, rarely of nails.

3. *Mode of transmission.* Direct contact with patient, and indirectly through toilet articles.

4. *Prevalence.* Rare in children in North America, and when occurring can usually be traced to immigrants from southern and eastern Europe.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic and cultural examinations of infected hairs or skin scrapings for *Trichophyton schoenleini (Achorion schoenleini).*

6. *Collection of specimens.* Submit hairs from infected areas or skin scrapings in sterile bottle in "MI" outfit. Mark history blank plainly "For fungi".

7. *Limitations of laboratory tests.* Suspected organisms must be identified by cultural characteristics.

FILARIASIS (MUMU)

1. *Etiologic agent.* A nematode worm. Several species of filariids are known to infect man; filariasis is most commonly caused by *Wuchereria bancrofti.*

2. *Source of infection.* The blood of an infected person.

3. *Mode of transmission.* In North America has been transmitted by the mosquito *Culex quinquefasciatus.* Other species of mosquitoes have been incriminated in other parts of the world. After the mosquito takes a blood meal from a person with circulating filaria embryos, the embryos develop in the mosquito into infective larvae in 14 to 21 days. Transmission is by the bite of the mosquito.

4. *Prevalence.* Not transmitted in the continental United States; previously reported cases were limited to Charleston, S. C. This focus of infection no longer exists. Common in certain tropical and sub-tropical parts of the world.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Microscopic examination of blood for microfilaria.

   B. Microscopic examination of fluid from lymph node puncture, hydrocele or similar material for microfilaria.

6. *Collection of specimens.*

   A. Collect thick blood films as for malaria examination. Use "MA" outfit. Follow directions given under "MALARIA". Late evening specimens are necessary and concentration methods may be advisable with filaria exhibiting periodicity; contact Bureau of Laboratories and a special outfit (containing 9 ml. of 2% formalin) will be provided. In either case plainly mark history blank "For filaria".

   B. Collect fluid in sterile bottle in "MI" outfit; mark history blank plainly "For filaria".

7. *Limitations of laboratory tests.* Identification of microfilaria is indication of infection. Since certain filaria exhibit periodicity both day and night bloods should be examined.

FOOD INFECTIONS (SALMONELLOSIS)

1. *Etiologic agent.* A variety of members of *Salmonella* group; most common in the United States are *Salmonella typhimurium (aertrycke), S. enteritidis,* and *S. choleraesuis (suipestifer).*

2. *Source of infection.* Usually animal sources. Rodents often infected, spreading infection through droppings; cattle and other livestock (including ducks and turkeys). Feces of patients and convalescent carriers; chronic carriers are rare.

3. *Mode of transmission.* Contamination of food through droppings of infected rodents; consumption of meat of infected animals, rarely ducks' eggs. Contamination of food or milk, through fecal contamination of hands of food-handlers.

4. *Prevalence.* Many cases of "common diarrhea" are in reality *Salmonella* infections; most commonly recognized following banquets or other meals of groups of persons; apparently less common in the United States than in Western Europe where many infections have been traced to meats and to ducks' eggs and fewer to rodents.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Cultural examinations of feces for *Salmonella* organisms including serological typing service.

   B. Cultural examination of suspected food vehicles for *Salmonella* organisms.

6. *Collection of specimens.* As soon as a case or an outbreak of food-borne gastroenteritis is suspected, consult your local health officer requesting an epidemiological investigation. This is desirable for the protection of the public. Meantime make certain that no food that may have been the vehicle of infection is discarded.

   A. Use "FE" outfit. Using the sterile swab in the outfit, transfer a portion of the stool about the size of a sphere 1/2" in diameter to the specimen jar and emulsify it in the glycerol solution. If stools are liquid, transfer to the specimen jar

with a clean spoon or other implement a quantity approximately 2 ml. in volume. Do not use spoon or other implement for more than one sampling unless carefully sterilized in the interim. Care should be taken that the stool is passed into a vessel that has been thoroughly disinfected or sterilized.

B. Rush suspected foods to the Bureau of Laboratories for examination; do not delay. Protect suspected foods from contamination during transit to Bureau of Laboratories. If possible, refrigerate food samples during transit.

7. *Limitations of laboratory tests.* In general, the *Salmonella* types reported refer to types of **animal** origin. Strictly speaking, the typhoid, para-A, para-B and para-C *(hirschfeldii)* organisms, all of **human** origin, are also members of the *Salmonella* group. No single type of *Salmonella* is constantly associated with a definite clinical syndrome since the illnesses produced may range from the subclinical case to a typhoidal syndrome and especially in infants may produce septicemia, meningitis or other grave illnesses involving any tissue or organ. However, the most frequent manifestations of *Salmonella* infection, particularly with the animal strains, are acute diarrheal conditions and acute gastroenteritis, food infections usually loosely spoken of as food poisoning. A food handler may spread infection if he himself is infected or becomes a carrier or if he dresses meat or fowl carrying the organisms without taking proper sanitary precautions. The *Salmonella* type reported is of importance in investigating the source of an illness or outbreak.

Where laboratory findings for *Salmonella* are negative, the possibility of staphylococcus food poisoning should be considered, particularly if the incubation period of the illness appears to have been 8 hours or less. See "FOOD POISONING: BACTERIAL INTOXICATIONS".

FOOD POISONING: BACTERIAL INTOXICATIONS

A. Staphylococcus

1. *Etiologic agent.* Toxin (enterotoxin) of certain strains of staphylococci. Toxin is stable at boiling temperature; staphylococci do not produce intestinal infection but multiply in food, producing a toxin which is cause of poisoning.

2. *Source of infection.* Not known in most cases, believed to be of human origin.

3. *Mode of transmission.* Most common vehicle is custard-filled pastry; processed meats, especially ham, responsible for some outbreaks; outbreaks reported due to milk from cows with specifically infected udders.

4. *Prevalence.* Widespread; probably the principal cause of acute "food poisoning".

5. *Current laboratory services. Available at Bureau of Laboratories:* Examinations of foods for enterotoxigenic staphylococci and for enterotoxin (Stone and Dolman tests).

6. *Collection of specimens.* Rush suspected foods to laboratory in suitable container; accompany by description of symptoms and probable incubation period.

7. *Limitations of laboratory tests.* The enterotoxin is preformed by bacterial growth in the foods. Hence, stool cultures are not recommended except when *Salmonella* infections must be excluded. In general, food poisoning occurring within 8 hours (usually 2-4 hours) after ingestion of the suspected food supports the probability of illness due to staphylococcus enterotoxin as opposed to *Salmonella* infections although relatively short incubation periods (6-12 hours) for the latter have been known. See "FOOD INFECTIONS (SALMONELLOSIS)".

   A negative test for enterotoxin may result when unaffected portions of the food are all that remain for sampling.

B. Botulinus (Botulism)

1. *Etiologic agent.* The toxin produced by the botulinus bacillus *Clostridium botulinum, (C. parabotulinum)* in foods improperly processed.

2. *Source of infection.* (Not an infection but a poisoning). Food usually taken uncooked from cans or jars not subjected to adequate heat of sufficient duration or under sufficient pressure during the processing.

3. *Mode of transmission.* Only by eating food containing the botulinus toxin.

4. *Prevalence.* Sporadic cases and groups of cases occur in all countries and always in relation to some perishable food product which has been so kept or preserved as to permit the development, under partially anaerobic conditions, of *Clostridium botulinum,* to the extent of forming the toxin that causes the symptoms. In the United States the disease has in recent years followed most commonly the use, without further or adequate cooking, of home-canned vegetable and meat products.

5. *Current laboratory services. Available at Bureau of Laboratories:* Examination for toxin of *Clostridium botulinum.*

6. *Collection of specimens.* Rush suspected foods to laboratory in suitable container; accompany by description of symptoms and probable incubation period; state if botulism is suspected.

7. *Limitations of laboratory tests.* Successful demonstration of the toxin is dependent upon selection of samples of food most likely to have supported the growth of *Clostridium botulinum,* an organism which grows only in the absence of air and does not usually grow in acid fruits and vegetables. Home-canned products are most likely sources.

GAS GANGRENE (ANAEROBIC WOUND INFECTIONS)

1. *Etiologic agent.* Various anaerobic spore-forming bacilli, *Clostridium perfringens* and many other species.

2. *Source of infection.* The organisms are widely distributed in nature but their main habitat is the soil. Some are common intestinal inhabitants of man and other animals.

3. *Mode of transmission.* By introduction of organisms into deep wounds at time of injury or by subsequent contamination.

4. *Prevalence.* Although the organisms are widespread in nature they require dead tissue and anaerobic conditions for growth and toxin production; hence the incidence is kept low by modern asepsis and surgical technic.

5. *Current laboratory services. Available at Bureau of Laboratories:* Bacteriological examinations of pus or curettings from wound.

6. *Collection of specimens.* Use "MI" outfit. Place material in sterile bottle in outfit. Mark history blank plainly "For anaerobic wound infection".

7. *Limitations of laboratory tests.* Only those spore-forming organisms which are obligatory anaerobes are significant.

GERMAN MEASLES (RUBELLA)

1. *Etiologic agent.* A filterable virus.
2. *Source of infection.* Secretions of the mouth and possibly the nose.
3. *Mode of transmission.* By direct contact with the patient or with articles freshly soiled with the discharges from the nose or throat of the patient.
4. *Prevalence.* Epidemic in expression, occurring mostly in childhood, but more in adults than is the case with measles; more prevalent in winter and spring than at other seasons.
5. *Current laboratory services.* No practicable diagnostic laboratory aid known to the Bureau of Laboratories.

GLANDERS

1. *Etiologic agent.* Glanders bacillus. *Malleomyces mallei (Bacillus mallei).*

2. *Source of infection.* Discharges from open lesions of mucous membranes or of the skin of infected horse or man (e. g., pus and mucus from the nose or throat, or bowel discharges).

3. *Mode of transmission.* Contact with infected horse or man or with articles freshly soiled by discharges therefrom.

4. *Prevalence.* Rare and sporadic and almost exclusively in men occupied about horses. In widespread and local epidemics as an epizootic in horses.

5. *Current laboratory services. Available at Bureau of Laboratories:* Cultural examinations of pus or exudates from lesions for *Malleomyces mallei.* Arrangements for serological tests of blood, particularly from animal sources, can be made when needed.

6. *Collection of specimens.* Contact Bureau of Laboratories before collecting specimens. Special instructions will be given.

7. *Limitations of laboratory tests.* Cultural identification of the organism is confirmatory of glanders suspected clinically. Serological tests are of value only in chronic disease.

GONORRHEA

1. *Etiologic agent.* Gonococcus, *Neisseria gonorrhoeae.*

2. *Source of infection.* Discharges from lesions of inflamed mucous membranes and glands of infected persons.

3. *Mode of transmission.* By direct personal contact with infected persons, and rarely by indirect contact with articles freshly soiled with the discharges of such persons. In adults by sexual intercourse; in children by personal and indirect contact with discharges; in the new born by ophthalmic infection at birth.

4. *Prevalence.* Widespread. Occurs among both sexes and at all ages, but is most prevalent among persons in the age groups of greatest sexual activity.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Microscopic examinations of smears.

   B. Cultural examination of pus or other exudate, and of urine in male patients.

   C. Complement-fixation tests as aids to the diagnosis of obscure conditions of possible gonococcal etiology (such as gonococcal arthritis) are available in limited numbers by arrangements with cooperating laboratories.

6. *Collection of specimens.*

   A. **Smears:**

   Use "GC" outfit. Make at least two smears. Mark or number each to indicate whether from urethra, cervix or other source. In the male take both smears from the urethral discharge (unless prostatic smears are preferred). In the female, take one from the urethral discharge, if there is any, and the other from the cervix uteri. Do not collect the discharge from between the labia or from the vagina as it is unsatisfactory for microscopic examination. In both sexes milk the urethra before making the smears.

   Make a thin smear from the discharge on one of the slides with one of the swabs, and a second smear on the other slide with the other swab. Let each smear dry at room temperature before putting the slides together. (Failure to observe

this precaution makes a reliable examination impossible.)

Diagnosis of specific vaginitis in children should always be confirmed by culture.

B. **Cultures:**

Use "NC" outfit which includes sterile swabs (set into corks and protected by tubes) and tubes of a sterile blood-dye mixture.

1. **General directions:** Remove from the tube the cork to which the swab is attached. Touch the swab to the exudate. Place it in a tube containing the blood-dye mixture and close the tube with the cork to which the swab is attached. When obtaining material from the cervix, use dressing forceps or a similar device for grasping the cork end of the swab.
2. **For male patients — acute cases:** Cleanse the glans penis with soap and water and collect the pus at the meatus with the sterile swab.
3. **For male patients — chronic cases and criterion of cure:** Cleanse the glans penis with soap and water and compress the anterior urethra between thumb and finger. Massage the prostrate with index finger of other hand. Release constriction of anterior urethra and collect secretion on swab.
4. **For female patients — urethral cultures:** Cleanse the os with a dry sterile cotton pledget and collect the urethral exudate on swab. Sufficient exudate may be obtained by applying pressure to Skene's ducts and to the urethra. In infants and children the swab should be inserted into the vagina and not merely brought into contact with the vulva.
5. **For female patients — cervical cultures:** Material is collected best with the aid of a speculum but **no lubricating jelly** should be used. Warm water is suggested. Remove cervical plug if present. Compress cervix to express glandular secretions in the deeper tissues and collect the exudate on the sterile swab.

6. After collection **return outfits as promptly as possible to laboratory.**

C. **Complement fixation test:**

Submit about 5 ml. blood in bottle in "MI" outfit. Mark history blank plainly "For GC complement fixation".

7. *Limitations of laboratory tests.*

A. **Smears:**

Results reported as positive are based on the finding of gram-negative intracellular diplococci morphologically identical with *Neisseria gonorrheae.* A positive result means that the microscopic picture is considered typical of that found in smears from persons infected with gonococci. Confirmation by culture should be made in vaginitis of children or whenever clinical findings are not indicative of infection.

If the smears are properly prepared, a negative result in suspected acute cases is fairly conclusive evidence of the absence of gonococci. In chronic cases it is of only limited value and should be confirmed by future smear examinations and by culture.

It is especially difficult to demonstrate intracellular gonococci in chronic gonorrhea of the female. Too much reliance should not be placed on any single negative laboratory examination.

It is generally considered good practice to submit both smears and cultures at intervals when judging the results of treatment.

B. **Cultures:**

A single negative specimen is not evidence of absence of specific infection. In general, greater reliability can be placed upon freshly cultured specimens than upon those delayed in transit to the laboratory. Periodic cultural examinations may be helpful in evaluating effectiveness of treatment.

C. **Complement-fixation tests:**

Complement fixation tests for gonococcal infection are of some value only in chronic conditions such as arthritis or endocarditis occurring as sequelae to possible gonococcal infections. A negative result is not conclusive. Complement fixation tests are of little value in acute gonorrhea.

GRANULOMA INGUINALE

1. *Etiologic agent.* Donovan body, *Klebsiella granulomatis.*

2. *Source of infection.* Discharges from lesions.

3. *Mode of transmission.* Direct contact by skin and mucous membranes during sexual intercourse with infected persons.

4. *Prevalence.* Widely prevalent in tropical and subtropical areas; endemic and recognized with increasing frequency in the United States.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic examination of scrapings from ulcers for Donovan bodies.

6. *Collection of specimens.* Take scrapings of material from ulcers and make smears on microscope slides contained in a "GC", "VI" or "MA" outfit. Mark history blank plainly "For Donovan bodies".

7. *Limitations of laboratory tests.* This disease is not to be confused with lymphogranuloma venereum (sometimes called lymphogranuloma inguinale). Laboratory findings are of value only to furnish presumptive evidence in support of clinical findings and history. Negative laboratory findings are not conclusive. According to some authorities, the Donovan bodies are bacteria similar to Friedlander's bacillus, *Klebsiella granulomatis,* but others consider these organisms to be secondary invaders.

HAVERHILL FEVER

1. *Etiologic agent.* Probably *Streptobacillus moniliformis (Haverhillia multiformis).*

2. *Source of infection.* Usually the bite of an infected rat; other possible sources not clearly delineated.

3. *Mode of transmission.* Contamination of wound at time of bite; possibly through ingestion of contaminated unpasteurized milk.

4. *Prevalence.* Sporadic; rare outbreaks.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Bacteriological examinations of pus or exudates from lesions.

   B. Blood cultures.

   C. Agglutination tests on blood specimens available through arrangement with cooperating laboratories.

6. *Collection of specimens.*

   A. Use "HS" outfit. Collect material from lesion on sterile swab provided and plunge into agar jelly before sending to laboratory. Mark history blank plainly "For Haverhill fever".

   B. Use "BC" outfit. Collect aseptically 5-10 ml. venous blood and introduce into medium in bottle directly from syringe through a sterile needle, preferably not the one used for venipuncture. The bottle cap has a special diaphragm through which the sterile needle is plunged to introduce the blood.

   C. Use "MI" outfit. Collect aseptically 5-10 ml. venous blood in sterile bottle provided. Mark history blank plainly "For Haverhill fever".

7. *Limitations of laboratory tests.* Isolation of the organism is confirmatory of the clinical syndrome. Agglutination tests of some value after first 10 days of illness.

HEMORRHAGIC JAUNDICE

(Icterohemorrhagic Spirochetosis, Weil's Disease)

1. *Etiologic agent.* *Leptospira icterohaemorrhagiae,* found in the blood or urine of patients and in the renal tract of rats. *L. canicola,* primarily a spirochete of dogs, is found in some human cases.
2. *Source of infection.* Urine and feces of rats, dogs, foxes, sheep, cats, and mice are at times involved. Wild rats often harbor leptospira in their kidneys. They are persistent carriers.
3. *Mode of transmission.* It appears that ingestion of contaminated food and water plays a role and that continued exposure of abraded or unabraded skin to alkaline waters containing leptospira may lead to infection. Sewer workers, retail and wholesale fish dealers, miners, and veterinarians are especially exposed to infection.
4. *Prevalence.* The disease is present in rats over the entire world. Sporadic human cases have been reported widely in the United States.
5. *Current laboratory services. Available at Bureau of Laboratories:*
   A. Animal inoculation tests.
   B. Agglutination tests.
6. *Collection of specimens.*
   A. Collect blood (about 5 ml.) for animal inoculation (during first 3-5 days of illness only) in sterile bottle in "MI" outfit. Indicate duration of disease on history blank. Plainly mark history blank "For Leptospira". Urine for animal inoculation should not be collected unless more than 5 days have elapsed from onset. Submit urine specimen in sterile bottle in "MI" outfit.
   B. Collect blood for agglutination test (only after first 10 days of illness) in sterile bottle in "MI" outfit.
7. *Limitations of laboratory tests.* *Leptospira* are sometimes difficult to demonstrate. Although direct examination of blood early in the disease by dark-field examination and by study of thick films is sometimes successful, animal inoculation tests on blood are more likely to be successful. Failure to demonstrate *Leptospira* or leptospiral agglutinins is presumptive, but not necessarily conclusive, evidence that jaundice observed clinically is due to other causes, including a virus.

HEPATITIS, INFECTIOUS (ACUTE CATARRHAL JAUNDICE)

1. *Etiologic agent.* A specific filterable virus.

2. *Source of infection.* Discharges from alimentary tract of infected persons and possibly also from the nose and mouth. The blood may contain the infectious agent. There may be carriers.

3. *Mode of transmission.* Unknown.

4. *Prevalence.* Epidemics are most commonly reported from rural areas and from institutions. Most outbreaks begin during the fall and winter months.

5. *Current laboratory services.* None available at Bureau of Laboratories except for exclusion of leptospiral infection (which may usually be ruled out more readily on clinical grounds). A few virus research laboratories have diagnostic facilities available.

6. *Collection of specimens.* Contact Bureau of Laboratories before submitting any specimens.

7. *Limitations of laboratory tests.* At present the lack of facilities for specific diagnostic aids constitutes the main limitation. Until such time as facilities for the study of virus infections are more generally available, recognition of this disease must remain a clinical problem.

HOOKWORM DISEASE (ANCYLOSTOMIASIS)

1. *Etiologic agents.* *Necator americanus* and *Ancylostoma duodenale.*

2. *Source of infection.* Usually soil contaminated with infective larvae from ova in stools deposited by infected persons. Larvae usually penetrate through the skin, although infection can take place by mouth.

3. *Mode of transmission.* The infective or third-stage larvae penetrate the skin, usually of the foot, and pass via the lymphatics to the inferior vena cava and the right heart, thence in the blood stream to the lungs, where they pierce the capillary walls and pass into the alveoli. They then pass up the bronchi and trachea to the throat, whence they are swallowed and finally reach the small intestine, where they develop to maturity. Infection can take place by mouth from water, soil, or contaminated objects harboring infected larvae; however, the chief mode of infection is through the skin.

4. *Prevalence.* Widely endemic in areas having favorable soil, moisture and temperature for development and where winter temperatures are not sufficiently low to destroy larvae in soil.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic examination of feces for ova (or larvae) of *Necator americanus* and *Ancylostoma duodenale.*

6. *Collection of specimens.* Use "PD" outfit. With a tongue depressor or similar implement take specimens of feces from different parts of the stool shortly after it is passed and place in bottle supplied with outfit.

7. *Limitations of laboratory tests.* Since hookworms do not multiply in the body, a light infection may not mean clinical hookworm disease. For the evaluation of treatment a series of negative stool examinations has more value than a single specimen.

HYDATID DISEASE (ECHINOCOCCOSIS)

1. *Etiologic agent.* ***Echinococcus granulosus (Taenia echinococcus).***

2. *Source of infection.* Infected dogs or other carnivores.

3. *Mode of transmission.* Ingestion of ova following contact with infected dogs or by contamination of food with ova from excreta of infected dogs.

4. *Prevalence.* Occurs throughout the world but is rare in North America.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Complement-fixation tests on blood specimens available through special arrangement with cooperating laboratories.

   B. Examinations of cyst fluid for scolices of *Echinococcus granulosus.* (Exploratory aspiration not recommended.)

6. *Collection of specimens.*

   A. Use "MI" outfit. Collect aseptically 5-10 ml. of blood and place in sterile bottle in outfit. Mark history blank plainly "For echinococcus".

   B. When cysts are encountered at autopsy or during operation, place fluid in bottle in "MI" outfit; mark history blank plainly "For echinococcus".

7. *Limitations of laboratory tests.*

   A. A positive complement-fixation test is confirmatory of clinical evidence. Negative results are not entirely conclusive.

   B. The examination of cyst fluid for scolices is of limited value since exploratory aspirations of the fluid is a dangerous procedure and securing of material is usually limited to autopsy specimens or specimens taken at operation.

IMPETIGO CONTAGIOSA

1. *Etiologic agent.* Probably staphylococci or streptococci.

2. *Source of infection.* Lesions on the skin of an infected person; possibly discharges from the nose and throat.

3. *Mode of transmission.* Directly by contact with the moist discharges of the skin lesions, or indirectly by contact with articles recently soiled by those discharges. The infection may be readily inoculated from place to place on the patient's body by scratching.

4. *Prevalence.* Common among children, especially in warm weather. Occurs sporadically and also in epidemic outbreaks in children's institutions and summer camps. Likely to spread rapidly where measures of personal hygiene are neglected and where skin lesions are frequent following scratching.

5. *Current laboratory services. Available at Bureau of Laboratories:* Cultural identification of organisms present in suspected lesions.

6. *Collection of specimens.* Use sterile swab in "HS" outfit for collection of pus or exudate from lesions from which crusts have been removed. Remove plug from tube enclosed in outfit and plunge swab after taking material into the agar in the bottom of tube. Swab must be left in that position and the plug inserted around other end of swab in the tube. Exercise aseptic precautions throughout. Return outfit to the laboratory by the quickest possible method. Delay in transit may result in false negative results. Mark history blank plainly "For identification of organism".

7. *Limitations of laboratory tests.* Bacteria present in skin lesions may occur as secondary invaders. In this disease, the appearance of the lesions is a greater diagnostic aid than cultures as the latter yield evidence of distinctly inferior value.

INFLUENZA

1. *Etiologic agent.* Two distinct types of virus, designated as Type A and Type B, have been identified. The more widespread recent epidemics have been associated with influenza A. Influenza B has usually been found in smaller and more localized outbreaks. In some epidemics neither Type A nor Type B has been found.

2. *Source of infection.* Probably discharges from the mouth and nose of infected persons and articles freshly soiled by such discharges.

3. *Mode of transmission.* Believed to be by direct contact, by droplet infection, or by articles freshly soiled with discharges of the nose and throat of infected persons.

4. *Prevalence.* Variable, in pandemics, local epidemics and as sporadic cases, often unrecognized by reason of indefinite clinical symptoms. In epidemics may affect up to 50 per cent of the population. Occurs pandemically at irregular intervals.

5. *Current laboratory services.* None available at present at Bureau of Laboratories. Certain virus laboratories are equipped to isolate the virus from nasopharyngeal washings and to perform both complement-fixation and neutralization tests on blood from patients and convalescents, but such tests are being done principally from the experimental viewpoint at present.

6. *Collection of specimens.* Contact Bureau of Laboratories before collecting any specimen. Instructions will be given providing it is feasible to enlist the services of a virus research laboratory.

7. *Limitations of laboratory tests.* At present the lack of facilities for specific diagnostic aids constitutes the main limitation. Demonstration of the virus or of complement-fixing or neutralizing antibodies is confirmatory of clinical observations and indicative of the type of virus present.

KERATO-CONJUNCTIVITIS, INFECTIOUS (SUPERFICIAL PUNCTATE KERATITIS, NUMMULAR KERATITIS)

1. *Etiologic agent.* Considered to be a specific filterable virus.

2. *Source of infection.* Probably the discharge from the eye of an infected person or a carrier.

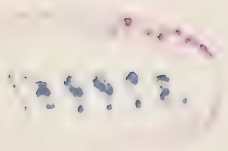
3. *Mode of transmission.* Apparently contact with an infected person or carrier or with articles freshly soiled with discharges of such person.

4. *Prevalence.* Occurs in epidemic form in warm climates, also among industrial employees in temperate climates involving a small percentage of the individuals in the groups affected.

5. *Current laboratory services.* None available at present at Bureau of Laboratories. Certain virus laboratories have done experimental work which shows promise of future application.

6. *Collection of specimens.* Contact Bureau of Laboratories when outbreak is suspected to determine whether or not laboratory examinations are feasible.

7. *Limitations of laboratory tests.* At present the lack of facilities for specific diagnostic aids constitutes the main limitation. Virus laboratories may be able to perform laboratory tests of diagnostic value.



LEISHMANIASIS (AMERICAN) MUCOCUTANEOUS LEISHMANIASIS, ESPUNDIA, UTA, BUBAS

1. *Etiologic agent.* *Leishmania braziliensis.*

2. *Source of infection.* The leishmania in cutaneous and mucocutaneous lesions.

3. *Mode of transmission.* Presumably through the bite of infected sand flies of the genus *Phlebotomus;* also by direct contact with infected individuals.

4. *Prevalence.* Reported from every country in South America except Chile. Reported from Central America and Mexico.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic examination of scrapings from suspected lesions.

6. *Collection of specimens.* Use "MA" outfit. Spread scrapings from suspected lesions on microscope slides and allow to dry. Mark history blank plainly "For *Leishmania*".

7. *Limitations of laboratory tests.* Identification of *Leishmania* in stained preparations is diagnostic of the disease when made by a specially trained observer.

LEPROSY

1. *Etiologic agent.* Leprosy bacillus, *Mycobacterium leprae.*

2. *Source of infection.* Discharges from lesions.

3. *Mode of transmission.* Intimate and prolonged contact with infected individuals and some other as yet undetermined factor are apparently necessary.

4. *Prevalence.* Endemic in some Gulf coast areas of the United States, Hawaii, Philippines, and Puerto Rico. Prevalence practically confined to tropical and subtropical areas. Usually more frequent among adolescent and young adult males.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic examination of smears from cutaneous lesions. No other laboratory aids to diagnosis have proved practicable.

6. *Collection of specimens.* Smear scrapings or exudates from cutaneous lesions on microscope slides contained in "MA" outfit. Mark history blank plainly "For leprosy".

7. *Limitations of laboratory tests.* Positive results indicate that acid-fast bacilli occurring in "lepra cells" or in pockets characteristic of *Mycobacterium leprae* have been observed. The organism is morphologically identical with that of tuberculosis. Cultures and animal inoculations are not feasible. There is no practicable laboratory aid to diagnosis of those types of leprous infection not complicated by cutaneous lesions.

LYMPHOGRANULOMA VENEREUM (INGUINALE) AND CLIMATIC BUBO

1. *Etiologic agent.* A specific filterable virus.
2. *Source of infection.* Discharges from lesions.
3. *Mode of transmission.* Direct contact by skin and mucous membranes, almost exclusively in sexual relations with infected persons, or indirectly by articles soiled with discharges from the lesions of such persons.
4. *Prevalence.* A common venereal infection among Negroes in the poorer sections of American cities. Widely prevalent in the tropics and common among inmates and clients of brothels in seaports.
5. *Current laboratory services. Available at Bureau of Laboratories:* Complement fixation tests.
6. *Collection of specimens.* Collect about 5 ml. of blood, using aseptic precautions, and place in sterile bottle in "MI" outfit. Allow to clot firmly. Store in refrigerator if held for any length of time before transmitting to laboratory. Mark history blank plainly "For lymphogranuloma venereum".
7. *Limitations of laboratory tests.* Complement fixation findings correlate with Frei test findings in 70-80% of cases. Negative complement fixation tests do not conclusively rule out this infection. Positive findings are known to occur in the absence of this infection when patient has cirrhosis of the liver or an infection with certain related viruses (psittacosis, virus pneumonias). Nevertheless, the test is a distinct aid in the confirmation of clinical findings.

MALARIA

1. *Etiologic agent.* The several species of microörganisms: *Plasmodium vivax* (tertian), *Plasmodium malariae* (quartan), *Plasmodium falciparum* (estivo-autumnal or malignant subtertian), *Plasmodium ovale.*

2. *Source of infection.* The blood of an infected individual.

3. *Mode of transmission.* By bite of infected anopheline mosquitoes. The mosquito is infected by biting an individual suffering from acute or chronic malaria. The parasite develops in the body of the mosquito for a variable period of time depending on the external temperature, under favorable conditions from 10 to 14 days (21 days for quartan), after which time the sporozoites appear in its salivary glands. The disease may be transmitted also by blood transfusion or by injecting whole human blood: also by common use of unsterilized hypodermic syringe (as by drug addicts).

4. *Prevalence.* Widespread in tropical and subtropical areas.

5. *Current laboratory services. Available at Bureau of Laboratories:* Identification of species of *Plasmodium* by microscopic examination of combined thick and thin blood films.

6. *Collection of specimens.* Preferably collect at least two specimens, one to be taken 12-24 hours after a chill and the other just before a chill is expected to recur. Each time collect both thick and thin blood films using slides in "MA" outfit, as follows:

   A. **Thick film:** Cleanse the ball of a finger or toe or the ear lobe with alcohol, allow to dry and puncture with a sterile lance or needle having a cutting edge. Avoid excessive bleeding but puncture deeply enough to produce a flow of blood with only slight pressure since bleeding produced by hard squeezing consists chiefly of serum which is unsatisfactory for reliable examination. The thick film should cover an area about the size of a dime. In judging the proper

thickness, ordinary printing can just be read through the center of the wet film, which should be several layers of erythrocytes thick in the center with thinner edge. Collect blood on slide in one of the following ways:

1. Touch under surface of slide about 1/4 inch from end opposite the etched portion to the large, rotund drop of blood on the punctured skin and without losing contact with the drop or touching the skin move the slide in narrow circles until a smear of satisfactory thickness and size is made.

2. Place 3-5 average drops of blood close together on the slide about 1/4 inch from the end opposite the etched portion and immediately puddle these into one homogeneous drop of proper size using a needle or the corner of a clean slide.

B. **Thin film:** After preparing the thick film place a small drop of blood on the slide near the etched portion (not on it) opposite the thick film. Place the narrow edge of another slide held at an angle of about 45° across this drop which will instantly spread all along the line of contact between the slides. Then, without pressure push the inclined slide forward toward but not into the thick film, leaving behind a thin, smooth blood film.

Allow both thick and thin films to dry in air with slides on a flat surface. Do not apply heat. Do not replace in slide holder until fully dry. Protect from flies or other insects.

7. *Limitations of laboratory tests.* Light infections may be missed, particularly with the thin film only. A few parasites may be found in the thick film only in such cases. Because of the fact that *Plasmodium falciparum* (aestivo-autumnal parasite) disappears from the peripheral blood soon after the chill, a series of examinations is more frequently necessary to detect this parasite than for detection of the other species. Proper identification of the parasites requires observation of the films by a specially trained microscopist.

MEASLES (RUBEOLA)

1. *Etiologic agent.* A specific filterable virus.

2. *Source of infection.* Buccal and nasal secretions of an infected individual.

3. *Mode of transmission.* Directly from person to person; indirectly through articles freshly soiled with the buccal and nasal discharges of an infected individual. One of the most easily transmitted of the communicable diseases.

4. *Prevalence.* Universal. Probably 80 to 90 per cent of all persons surviving to the twentieth year of life have had an attack, and rarely does a person go through life without having had measles. Occurs most commonly in children between 5 and 14 years of age, but many cases are in children under 5. Endemic in large population areas.

5. *Current laboratory services.* No practicable laboratory aid to diagnosis known to Bureau of Laboratories.

MENINGOCOCCUS MENINGITIS (CEREBROSPINAL FEVER),* MENINGOCOCCEMIA

1. *Etiologic agent.* Meningococcus, or *Neisseria meningitidis (N. intracellularis).* Four main serologic types or groups are recognized. Group I has been more frequently found in recent epidemics in the United States.

2. *Source of infection.* Discharges from the nose and throat of patients or carriers, as the organisms are commonly carried in the nasopharynx. Carrier prevalence of 25 per cent or higher may exist without the occurrence of cases. During epidemic periods more than half of a military organization may be healthy carriers of the strain of meningococci responsible for the epidemic.

3. *Mode of transmission.* By contact with infected persons, that is, sick persons or carriers. Indirect transmission may perhaps occur through contact with articles freshly soiled with discharges from the respiratory tract of infected persons.

4. *Prevalence.* Endemic and epidemic. There are no limits in geographic distribution. Sporadic cases occur throughout the year in both urban and rural areas with the greatest incidence during the winter and spring. The disease exhibits high incidence at irregular intervals. The epidemic wave lasts usually 2 to 3 years.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Microscopic and cultural examination of spinal fluid from cases of meningitis.

   B. Blood culture service in cases of meningococcemia.

6. *Collection of specimens.*

   A. Collect aseptically about 10 ml. spinal fluid in

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* The laboratory services described apply equally as well to other bacterial meningitis.

the sterile bottle supplied in the "SF" outfit. Check examinations desired on history blank. **Rush to laboratory.**

B. Use "BC" outfit. Collect venous blood for culture aseptically in sterile syringe of 5-10 ml. capacity. Preferably replace used needle with another unused sterile one. Plunge needle through thin rubber top of stopper (previously swabbed with alcohol) and expel contents of syringe into medium in bottle. Rush to laboratory.

7. *Limitations of laboratory tests.* The laboratory procedures outlined above will detect infections other than meningococcic but animal inoculation tests on spinal fluid may be necessary if tuberculous meningitis is suspected. Cell counts (preferably made locally), total protein determinations and colloidal gold tests on spinal fluid may be valuable aids. The causative organism, *Neisseria intracellularis* (meningococcus), does not remain viable for long periods of time outside the body and it is therefore necessary to rush culture material to the laboratory as soon as possible. For this and other reasons, nasopharyngeal cultures for the detection of suspected carriers are not feasible services for a central laboratory.

MONONUCLEOSIS, INFECTIOUS (GLANDULAR FEVER)

1. *Etiologic agent.* Unknown.

2. *Source of infection.* Probably discharges from the nose and throat of infected persons.

3. *Mode of transmission.* Direct contact with infected persons. The importance of articles soiled with discharges of infected persons is undetermined.

4. *Prevalence.* Observed in many parts of the world and is probably much more prevalent and more widely distributed than indicated by reported incidence. Epidemics are most frequently recognized in schools and children's institutions; the recognized incidence is comparatively high among medical students, nurses, hospital personnel and among other groups having access to medical services where blood examinations are made routinely.

5. *Current laboratory services. Available at Bureau of Laboratories:* Serological examination of blood for heterophile agglutinins specific for infectious mononucleosis.

6. *Collection of specimens.* Collect 5-7 ml. venous blood aseptically and place in sterile bottle in "MI" outfit. Mark history blank plainly "For infectious mononucleosis".

7. *Limitations of laboratory tests.* Sheep red cell agglutinins may be present in the blood either naturally (native antibodies) or as a result of serum therapy (serum sickness) as well as in infectious mononucleosis. Since the simple agglutination test (Paul-Bunnell test) does not differentiate among these conditions, it is necessary to adsorb the serum with guinea-pig kidney before positive results of the test can be considered specific for infectious mononucleosis. This adsorption removes all other heterophile antibodies. Examination of a blood smear may provide evidence of the disease in very early cases or in those which do not develop the characteristic heterophile agglutinins.

MUMPS (INFECTIOUS PAROTITIS)

1. *Etiologic agent.* A specific filterable virus.

2. *Source of infection.* Secretions of the mouth and possibly of the nose.

3. *Mode of transmission.* By direct contact with an infected person or with articles freshly soiled with the discharges from the nose and throat of such infected persons.

4. *Prevalence.* This disease is decidedly less prevalent than the other common communicable diseases of childhood such as measles, whooping cough and chickenpox. Winter and spring are the seasons of greatest prevalence. Its occurrence is sporadic and epidemic except in large cities, where it is endemic. Outbreaks occur more frequently and are of a more serious character in aggregations of young people, especially under conditions of military mobilization.

5. *Current laboratory services.* No practicable laboratory aid to diagnosis known to Bureau of Laboratories. Successful use of a complement fixation test has been reported but is still in the experimental stage.

MYCOTIC INFECTIONS, MISCELLANEOUS (INCLUDING BLASTOMYCOSIS, MONILIASIS, TORULOSIS, THRUSH)

1. *Etiologic agent.* Among others are *Monilia albicans (Monilia candida), Blastomyces dermatitidis, Cryptococcus hominis (Torula histolytica).*

2. *Source of infection.* The organisms probably are widely distributed in nature but infection may occur from contact with such sources as infected animals.

3. *Mode of transmission.* By contact with infected materials or animals; trauma may be a contributing factor.

4. *Prevalence.* Relatively infrequent but worldwide in distribution.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic and cultural examinations of body fluids, exudates, etc.

6. *Collection of specimens.* Collect body fluids, pus or other material from affected areas in sterile bottle in "MI" outfit; when lesion is small, collect material on sterile swab in "HS" outfit and plunge into agar jelly in tube furnished before returning to laboratory. Mark history blank plainly "For fungi".

7. *Limitations of laboratory tests.* Cultural identification of known pathogenic types is desirable in all cases because of the widespread distribution of fungi in nature.

PARATYPHOID FEVER

1. *Etiologic agent.* Paratyphoid bacillus A, B, or C; *Salmonella paratyphi, Salmonella schottmuelleri, Salmonella hirschfeldii.*

2. *Source of infection.* Bowel discharges and urine of patients or carriers and food, water, or milk contaminated with such discharges of infected persons. Healthy carriers may be numerous in an outbreak.

3. *Mode of transmission.* Conveyance of paratyphoid bacilli by direct or indirect contact with patient or carrier. Among indirect means of transmission are contaminated food, water, milk, and shellfish, and, under some conditions, flies.

4. *Prevalence.* Frequency has fallen with that of typhoid fever until in most parts of North America it is relatively rare, occurring sporadically or in small local carrier or contact epidemics, though probably more common than recognized due to the large number of unrecognized infections. Paratyphoid A fever less common than B.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Blood cultures (1-7 days' duration of disease).

   B. Agglutination tests (after 10th day of illness). Cultures are made from the clots after the serum has been removed.

   C. Feces, urine and bile cultures for isolation and typing of organism.

6. *Collection of specimens.*

   A. **Blood cultures:** Use "BC" outfit. Collect aseptically venous blood for culture in sterile syringe of 5-10 ml. capacity. Preferably replace needle used with another unused sterile one. Plunge needle through thin rubber top of stopper (previously swabbed with alcohol) and expel contents of syringe into medium in bottle. Rush to laboratory.

   B. **Agglutination tests and clot cultures:** Collect 5-10 ml. venous blood aseptically in sterile bottle in "TY" outfit. Allow to clot firmly before sending to laboratory.

C. **Feces:** Use "FE" outfit. Using the sterile swab in the outfit, transfer a portion of the stool about the size of a sphere 1/2 inch in diameter to the specimen jar and emulsify it in the glycerol solution. If stools are liquid, transfer to the specimen jar with a clean spoon or other implement a quantity approximately 2 ml. in volume. Do not use spoon or other implement for more than one sampling unless carefully sterilized in the interim. Care should be taken that the stool is passed into a vessel that has been thoroughly disinfected or sterilized.

Urine: Use "UR" outfit. Have patient void urine into previously sterilized vessel and place 10-15 ml. in sterile bottle provided in outfit.

Bile: Collect this material ordinarily only when carrier has been hospitalized for release from carrier state according to the requirements of the Sanitary Code of Connecticut. Use duodenal tube and place 10-15 ml. of biliary drainage in sterile bottle in "FE" outfit. Be sure to check history blank to indicate specimen is bile.

7. *Limitations of laboratory tests.*

A. For early diagnosis a blood culture is the best diagnostic aid. It should be taken during the first five days of illness, after which the organisms usually disappear from the blood stream. Isolation of a properly typed *Salmonella* establishes the cause of illness.

B. Agglutinins do not appear in the blood until 10-14 days after onset. The titer reaches its height during the third week of illness. Agglutinins produced as a result of previous infection or of prophylactic vaccination may persist for an indefinite period of time. Therefore, the presence of agglutinins is not diagnostic of itself and must be considered in the light of clinical findings and of past history of the patient. Since some patients may not develop agglu-

tinins, negative results are only suggestive, not conclusive, of absence of this disease.

Results of clot cultures have the same significance as those on blood cultures (See A. above). Negative results are, however, likely to occur after the first five days of illness.

C. Isolation of a properly typed *Salmonella* from the feces, urine or bile may mean the patient is a temporary or chronic carrier or that the organism isolated is the cause of the illness. Hence, the significance of positive results must be considered in the light of clinical findings. For release of cases and carriers, see the requirements of the Sanitary Code of Connecticut.

PEDICULOSIS (LOUSINESS)

1. *Infesting agent.* Head louse or body louse *(Pediculus humanus),* and crab louse *(Phthirus pubis).*

2. *Source of infestation.* Usually the hairy part of an infested person or, in the case of the body louse, the clothing of such a person.

3. *Mode of transmission.* Direct contact with an infested person and indirectly by contact with clothing and headgear of such persons.

4. *Prevalence.* Universal where there is neglect of washing of the person and the body clothing.

5. *Current laboratory services.* The diagnosis of this condition is not a laboratory function but identification of species of lice found, when there is some doubt of their identity, can be arranged through Bureau of Laboratories.

6. *Collection of specimens.* Place lice in alcohol or 10% formalin in any convenient tightly stoppered vial.

7. *Limitations of laboratory tests.* Occasionally other lice or mites, such as chicken lice, may be mistaken for head or body lice. Examination under a low-power lens will bring out distinguishing characteristics.

PEMPHIGUS NEONATORUM (IMPETIGO OF THE NEW BORN)

1. *Etiologic agents.* Probably staphylococci or streptococci.
2. *Source of infection.* Infected infants, attendants, or visitors.
3. *Mode of transmission.* By direct or indirect contact with infected persons or articles contaminated by them.
4. *Prevalence.* Occurs occasionally in nursery wards. Likely to spread rapidly.
5. *Current laboratory services. Available at Bureau of Laboratories:* Cultural identification of organisms present in suspected lesions.
6. *Collection of specimens.* Use sterile swab in "HS" outfit for collection of pus or exudate from lesions from which crusts have been removed. Remove plug from tube enclosed in outfit and plunge swab after taking material into the agar in the bottom of tube. Swab must be left in that position and the plug inserted around other end of swab in the tube. Exercise aseptic precautions throughout. Return outfit to laboratory by the quickest possible method. Delay in transit may result in false negative results. Mark history blank plainly "For identification of organism".
7. *Limitations of laboratory tests.* Bacteria present in skin lesions may occur as secondary invaders. In this disease, the appearance of the lesions is a greater diagnostic aid than cultures as the latter yield evidence of distinctly inferior value.

PERTUSSIS (WHOOPING COUGH)

1. *Etiologic agent.* Pertussis bacillus of Bordet and Gengou, *Hemophilus pertussis.*

2. *Source of infection.* Discharges from the laryngeal and bronchial mucous membranes of infected persons.

3. *Mode of transmission.* Contact with an infected person, or with articles freshly soiled with the discharges of such person.

4. *Prevalence.* Very prevalent, and a common disease among children everywhere regardless of race, climate, or geographic location. Although approximately 15 per cent of the cases occur in children under 2 years of age, 85 per cent of the deaths occur in this age group. Seasonal incidence variable, but mortality higher usually in spring months in North America. Cyclical occurrence irregular.

5. *Current laboratory services.* Cough plate cultures are feasible under certain conditions but are not routinely made at Bureau of Laboratories. Since this service cannot be extended to physicians on a statewide basis, only unusual circumstances justify examination of specimens. Agglutination tests for bedside use by physicians are used by some; materials may be purchased through certain biologics manufacturers.

6. *Collection of specimens.* Contact Bureau of Laboratories before submitting any specimens for this disease.

7. *Limitations of laboratory tests.* The causative organism, *Hemophilus pertussis,* will grow only upon specially prepared media and exposed cough plates must be placed under incubation immediately. A single negative culture is not conclusive. A positive agglutination test may be found during the second or third week following the onset of the first indefinite symptoms.

PIN WORM INFECTION (ENTEROBIASIS, OXYURIASIS)

1. *Etiologic agent.* The pinworm or seatworm, *Enterobius vermicularis (Oxyuris vermicularis).*

2. *Source of infection.* The infected human.

3. *Mode of transmission.* Direct by swallowing ova in contaminated food or drink. Fly contamination of food is a factor. Auto-infection is common.

4. *Prevalence.* Widespread. The incidence may be very high in certain groups especially children.

5. *Current laboratory services. Available at Bureau of Laboratories:* Examination of anal swabbings for ova of *Enterobius (Oxyuris) vermicularis*; identification of adult worms passed in feces.

6. *Collection of specimens.* Use "PW" outfit. Collect specimens in the morning only and immediately after patient has awakened. To collect eggs deposited by worms during patient's sleep, use cellophane swab on the end of the glass rod furnished in the tube by gently stroking the anus and adjacent area; return swab to tube and submit to the laboratory. For submission of adult worms use "PD" outfit.

7. *Limitations of laboratory tests.* Examination of stool specimens is not so reliable as examination of anal swabbings collected as described above because the eggs are actually deposited outside the anus by the gravid female of *Enterobius.*

PLAGUE

1. *Etiologic agent.* Plague bacillus, *Pasteurella pestis.*

2. *Source of infection.* Blood of infected rodents and, in the pneumonic form, the sputum of human cases. The primary or indigenous source of the disease is the so-called "sylvatic plague", the animal reservoir among such rodents as the tarbigan of Manchuria, and the ground squirrel and other rodents of the United States. Infection may reach man from these sources, or more often through the medium of the rat.

3. *Mode of transmission.* Direct in the pneumonic form. In other forms the disease is generally transmitted by the bites of fleas (*Xenopsylla cheopis* and certain other species), by which the disease is carried from rats to man, also by fleas from other rodents. Accidental, by inoculation.

4. *Prevalence.* Very rare in North America and insular possessions, and only sporadic cases, from exposure to infection in ground squirrels and other rodents west of the Mississippi. Focally distributed in various parts of the world.

5. *Current laboratory services.* Cultural examination of material aspirated from buboes, or of sputum in pneumonic type, can be made through special arrangements with Bureau of Laboratories.

6. *Collection of specimens.* Contact Bureau of Laboratories before collecting any specimens. Special instructions will be given.

7. *Limitations of laboratory tests.* For precise identification of the causative organism, *Pasteurella pestis,* animal inoculations and serological studies are required.

PNEUMONIA

A. Pneumococcal — Acute Lobar Pneumonia

1. *Etiologic agent.* Pneumococci Types I to XXXII inclusive account for about 95 percent of the cases, the remaining are due to the more rarely recognized types. *Streptococcus hemolyticus* produces an atypical pneumonia, interstitial in type, which may be confused with lobar pneumonia.

2. *Source of infection.* Probably discharges from the mouth and nose of infected persons and articles freshly soiled with such discharges.

3. *Mode of transmission.* By direct contact with infected person, or with articles freshly soiled with the discharges of the nose and throat of such person, and possibly from minute suspended particles containing the etiologic agent. Incidence of carriers is much higher than that of cases.

4. *Prevalence.* Common, and affecting at one time or other, between adolescence and old age, a large proportion of the population. No race or color and neither sex is exempt from likelihood of having this disease. Occurs in all climates and seasons, but most often in winter and spring and in regions where cold, windy, changeable, and inclement weather prevails. Occurs in epidemic form, particularly in institutions for adults.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Isolation and typing of organism.

   B. Blood cultures for *Diplococcus pneumoniae* (pneumococcus).

   C. Cultural examination and typing of pneumococci isolated from body fluids or discharges when such complications or sequelae as otitis media or meningitis occur.

6. *Collection of specimens.*

   A. **Sputum for typing:** Use "PN" outfit. Collect sputum which has been coughed up from the lungs. Avoid inclusion of saliva and superficial mucous secretions. Particularly include portions of sputum that are blood tinged. Place sputum

in sterile bottle in outfit. On young children from whom sputum is difficult to obtain, material coughed up from the lungs may be obtained directly on a good sized, sterile swab held to the back of the mouth when coughing occurs or is induced. The handle of the swab may be broken off and the swab placed in the sterile bottle provided in the outfit.

B. **Blood Cultures**: Use "BC" outfit. Collect aseptically venous blood for culture in sterile syringe of 5-10 ml. capacity. Preferably replace needle used with another unused sterile one. Plunge needle through thin rubber stopper (previously swabbed with alcohol) and expel contents of syringe into medium in bottle. Rush to laboratory.

C. Use "MI" outfit for body fluids; use "HS" outfit for pus or other discharges. Use aseptic technique. Mark history blank "For type of organism". Rush to laboratory.

7. *Limitations of laboratory tests.*

A. **Sputum typing**: Provided sputum is obtained from the deeper air passages, a negative specimen from individuals not treated with sulfa drugs is presumptive evidence of absence of pneumococcic infection.

The type of pneumococcus found in the sputum is considered of diagnostic significance in the absence of blood culture findings when symptoms are those of lobar pneumonia. Blood cultures should be taken to detect bacteremia and to follow results of treatment.

Other organisms such as *Klebsiella pneumoniae* (Friedlander's bacillus) and pyogenic cocci may produce pneumonia. *Mycobacterium tuberculosis* (tubercle bacillus) is also reported when found.

B. **Blood cultures**: A pneumococcus type isolated from blood culture is considered proof of etiological significance. Bacteriemia may not occur except in severe cases.

C. **Cultures of body fluids and discharges**: The type of organism reported may be considered of significance unless introduced in the collection of the specimen.

B. Bacterial Pneumonia, other than Pneumococcal

1. *Etiologic agent.* Various pathogenic bacteria of the mouth, nose, and throat, as the streptococcus, staphylococcus, *Klebsiella pneumoniae* and *Hemophilus influenzae.*

2. *Source of infection.* Probably discharges from the mouth and nose of an infected person, or articles soiled with such discharges.

3. *Mode of transmission.* By direct contact with infected person or with articles freshly soiled with discharges of nose or throat of such person.

4. *Prevalence.* Common only during prevalence of epidemic influenza or of other respiratory infections.

5. *Current laboratory services. Available at Bureau of Laboratories:* Cultural identification of organisms in sputum.

6. *Collection of specimens.* Use "PN" outfit. Collect sputum which has been coughed up from the lungs. Avoid inclusion of saliva and superficial mucous secretions. Particularly include portions of sputum that are blood tinged. Place sputum in sterile bottle in outfit. On young children from whom sputum is difficult to obtain, material coughed up from the lungs may be obtained directly on a good sized, sterile swab held to the back of the mouth when coughing occurs or is induced. The handle of the swab may be broken off and the swab placed in a sterile bottle provided in the outfit.

7. *Limitations of laboratory tests.* Organisms other than the pneumococcus most likely to be of significance when found in pneumonic sputum are: *Klebsiella pneumoniae* (Friedlander's bacillus), beta hemolytic streptococci and sometimes *Hemophilus influenzae* or the pyogenic cocci. The presence of these organisms in sputum constitutes contributory evidence only and is not diagnostic.

C. Primary Atypical Pneumonia

1. *Etiologic agent.* The causative agent of the majority of atypical pneumonias is probably a virus.

2. *Source of infection.* Probably discharges from the mouth and nose of infected persons or articles freshly soiled with such discharges.

3. *Mode of transmission.* By direct contact with infected person or with articles freshly soiled with discharges of nose and throat of such person. Mild, unrecognized infections may play a role in the spread of the disease.

4. *Prevalence.* Occurs endemically and in epidemics at all seasons. Incidence is variable. In outbreaks in army camps attack rates of from 1 to 6 per cent of the troops per year have been reported. Similar attack rates are reported in civilian hospitals and institutions. Occurs in both sexes and at all ages, but is more frequent in adolescents and young adults.

5. *Current laboratory services.* None available at present at Bureau of Laboratories except those which aid in the exclusion of bacterial agents (See previous sections). Experimental studies in virus laboratories are not yet sufficiently well developed for practicable use.

6. *Collection of specimens.* For examinations to aid in exclusion of bacterial pathogens, use "PN" outfit. Collect sputum which has been coughed up from the lungs. Avoid inclusion of saliva and superficial mucous secretions. Particularly include portions of sputum that are blood tinged. Place sputum in sterile bottle in outfit. On young children from whom sputum is difficult to obtain, material coughed up from the lungs may be obtained directly on a good sized, sterile swab held to the back of the mouth when coughing occurs or is induced. The handle of the swab may be broken off and the swab placed in a sterile bottle provided in the outfit.

7. *Limitations of laboratory tests.* Results of examinations for the exclusion of bacterial pathogens are of indirect value only in the establishment of a diagnosis of primary atypical pneumonia as defined above.

POLIOMYELITIS

1. *Etiologic agent.* A specific filterable virus.

2. *Source of infection.* Nose and throat discharges of infected persons, more frequently those not suffering from a clinically recognized attack of the disease. Bowel discharges contain the virus.

3. *Mode of transmission.* The virus probably enters the body by way of the nose or mouth, presumably from a carrier or a person with a subclinical infection in most instances. Though the virus has been found in flies subject to fecal contamination, there is no good evidence of insects serving as vectors. Reliable evidence of spread by water supply is lacking.

4. *Prevalence.* Infection occurs practically throughout the world, but cases are most frequent in the cooler part of the temperate zone, occurring both sporadically and in epidemics at irregular intervals, with the highest incidence in late summer and fall. In northern United States an annual incidence of 10 per 100,000 population is ordinary.

5. *Current laboratory services.* None available at Bureau of Laboratories except when routine examinations of spinal fluid for total protein and colloidal gold test are desired. (Certain virus laboratories about the country have facilities for isolation of the virus for experimental purposes.)

6. *Collection of specimens.* Spinal fluid for routine examination may be submitted in "SF" outfit. Collect aseptically about 10 ml. of fluid in the sterile bottle in the outfit.

7. *Limitations of laboratory tests.* Cell counts of spinal fluid, preferably made locally, may aid in making an early diagnosis. Total protein and colloidal gold reactions provide indirect information of limited value.

PROTOZOAN INFECTIONS, MISCELLANEOUS

1. *Etiologic agent.* Among others, *Balantidium coli, Giardia lamblia, Chilomastix mesnili, Trichomonas vaginalis.*

2. *Source of infection.* Infected humans.

3. *Mode of transmission.* Direct contact or by articles or food soiled by infected persons. Flies may be an intermediary.

4. *Prevalence.* Widespread, usually sporadic but may occur in outbreak form especially in institutions.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic examinations of excreta or other material for type of organism.

6. *Collection of specimens.* Feces or urine may be submitted in bottle in "PD" outfit. Vaginal smears for *Trichomonas vaginalis* may be submitted on microscope slides in "VI" outfit provided history blank is plainly marked "For *Trichomonas*".

7. *Limitations of laboratory tests. Balantidium coli* is pathogenic. Species of *Giardia, Chilomastix* and *Trichomonas* are of doubtful pathogenicity except that the last may cause vaginitis.

PSITTACOSIS

1. *Etiologic agent.* A specific filterable virus.

2. *Source of infection.* Parrots, parakeets, love birds, canaries, pigeons, and other birds. Birds which are apparently well occasionally transmit the infection.

3. *Mode of transmission.* Contact with infected birds or their recent surroundings. Occasionally through a human case.

4. *Prevalence.* Usually in sudden house outbreaks among persons exposed to ill tropical birds. Deaths mainly confined to persons over 30 years of age. Milder cases not infrequent from slight exposure to pigeons or other birds not necessarily ill.

5. *Current laboratory services.* None available at present at Bureau of Laboratories. Special examinations can be arranged through cooperating virus laboratories.

6. *Collection of specimens.* Contact Bureau of Laboratories before submitting any specimens. Special instructions will be given.

7. *Limitations of laboratory tests.* Identification of the virus by animal inoculation tests is of diagnostic significance. Development of neutralizing antibodies in the blood during convalescence is strong presumptive evidence of infection.

RABIES

1. *Etiologic agent.* A specific filterable virus.

2. *Source of infection.* Infected animals, chiefly dogs; vampire bats in limited areas, notably Trinidad.

3. *Mode of transmission.* Usually bites by a rabid animal, occasionally through contact of such animal's saliva with scratch or other break in a person's skin. The milk or meat of infected animals, such as cows, is not dangerous for human use.

4. *Prevalence.* Rare in man. Prevalent on all continents except Australia. More prevalent among dogs and sometimes in wild carnivorous animals.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. **Animal rabies:** Microscopic examination of brain tissue for Negri bodies and animal inoculation tests for confirmation of negative results.

   B. **Human rabies:** Microscopic examination of brain tissue removed at autopsy and confirmatory animal inoculation tests.

6. *Collection of specimens.*

   A. **Animal heads:** Do not kill suspected animals unnecessarily. When clinical symptoms of the animal are suspicious begin antirabic treatment of persons bitten or otherwise exposed without waiting for a laboratory test. Keep suspected animals under observation according to instructions from your local health officer. A rabid animal does not recover; the disease is progressive and fatal. When suspected animal has died or has had to be killed, remove head and place in a metal container with a tightly fitting metal top. Pack this in ice before submission to the laboratory.

   B. **Human brains:** When permission for autopsy has been secured, place brain in suitable water-

tight container and pack in ice before submission to the laboratory.

7. *Limitations of laboratory tests.* The finding of typical Negri bodies in nerve cells of the brain is considered diagnostic of rabies. Animal inoculation tests complete the evidence.

Brain tissue may not contain Negri bodies in detectable numbers early in the disease. Negri bodies may not be found if those portions of the brain where they are usually found have been destroyed by gunshot or violent blow.

Decomposed brains are unreliable for examination and frequently unsuitable for animal inoculation tests.

RAT-BITE FEVER (SODOKU)*

1. *Etiologic agent.* *Spirillum minus (Spirochaeta morsus-muris).*

2. *Source of infection.* Usually bite of the rat; rarely cat, weasel, ferret, dog, or bandicoot.

3. *Mode of transmission.* During the bite, some of the animal's blood escapes from the injured or diseased buccal mucosa into the wound, or the conjunctival secretion of the rat may contaminate the wound. Blood from an animal in the laboratory may infect man.

4. *Prevalence.* Distribution is world-wide. Rare in North and South America and in most European countries; more common in Japan and in the Far East.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Animal inoculation tests of blood or of exudate from primary lesion for demonstration of the causative organism, *Spirillum minus.*

   B. Agglutination tests on blood for infection with *Streptobacillus moniliformis* are made through arrangements with cooperating laboratories since that infection (Haverhill fever) also may result from rat bites.

6. *Collection of specimens.* At height of fever collect 5-10 ml. of venous blood in sterile bottle in "MI" outfit. Mark history blank plainly "For rat-bite fever". Exudate from primary lesion may be collected on swab in "HS" outfit and must be rushed to laboratory.

7. *Limitations of laboratory tests.* A single negative result on a blood specimen is not conclusive. Therefore, examination of three blood specimens taken on successive days should be planned. Since laboratory animals are frequently parasitized by organisms which may be mistaken for *Spirillum minus*, positive animal inoculation tests must be confirmed by an experienced observer.

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* Another disease, Haverhill fever or infection with *Streptobacillus moniliformis (Haverhillia multiformis)* is also in its sporadic occurrence transmitted by the bite of an infected rat and appears to be more frequent in some parts of the United States than Sodoku (true rat-bite fever). It is usually characterized by arthritic symptoms and the eruption is not plague like. Localized epidemics may occur from unpasteurized milk. See page 43.

RELAPSING FEVER

A. Louse-Borne

1. *Etiologic agent.* A spirochete, *Borrelia recurrentis.*

2. *Source of infection.* The natural reservoir of infection is unknown. After biting an infected human being, lice *(Pediculus humanus)* become infected in about 16 days and remain so for life (30 or 40 days). Hereditary transmission in lice through the egg to the larval form is reported but has rarely been observed.

3. *Mode of transmission.* Transmission from man to man is effected by crushing an infected louse into the bite-wound or into an abrasion on the skin, or by rubbing louse feces or coxal fluid into an abrasion of the skin.

4. *Prevalence.* The disease is prevalent among primitive people who are louse infested. It is found in limited localities in Europe, Asia, North and South Africa and Central America. For more than a quarter of a century louse-borne relapsing fever has not been observed in the United States.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Dark-field examinations and animal inoculation tests on blood specimens.

   B. Examination of thick blood films available.

6. *Collection of specimens.*

   A. **Whole blood**: During febrile paroxysm collect 15 ml. venous blood aseptically and place in sterile bottle in "MI" outfit. Mark history blank plainly "For relapsing fever".

   B. **Blood films**: During febrile paroxysm make thick blood film. (See directions under "MALARIA".) Allow to dry in air without application of heat before sending to laboratory.

7. *Limitations of laboratory tests.* It is not possible to distinguish between species of *Borrelia* by ordinary laboratory examinations. Presence of any *Borrelia* in the blood is, however, confirmatory of the clinical syndrome of relapsing fever. Specimens taken during afebrile periods are likely to yield negative results

B. Tick-Borne

1. *Etiologic agent.* A spirochete, *Borrelia duttoni.*

2. *Source of infection.* Primarily an infection of wild rodents, transmitted by the genus of ticks *Ornithodorus.* In the United States *O. turicata* is a known vector in Texas and Kansas; *O. hermsi* in California, Colorado and Idaho. *O. talaje* is a vector in Panama, Central and South America, while *O. moubata* is the vector in tropical Africa.

3. *Mode of transmission.* Man is accidentally infected by a tick bite.

4. *Prevalence.* Widespread throughout tropical Africa. Foci have been observed in Spain, North Africa, Arabia, Iran, India, and other parts of Central Asia as well as in North and South America. In the United States human cases of tick-borne relapsing fever have been found to originate in limited localities of 11 different states — Colorado, California, Texas, Arizona, Kansas, Washington, Utah, Nevada, Idaho, New Mexico, and Oklahoma.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Dark-field examinations and animal inoculation tests on blood specimens.

   B. Examination of thick blood films.

6. *Collection of specimens.*

   A. **Whole blood:** During febrile paroxysm collect 15 ml. venous blood aseptically and place in sterile bottle in "MI" outfit. Mark history blank plainly "For relapsing fever".

   B. **Blood films:** During febrile paroxysm make thick blood film. (See directions under "MALARIA.") Allow to dry in air without application of heat before sending to laboratory.

7. *Limitations of laboratory tests.* It is not possible to distinguish between species of *Borrelia* by ordinary laboratory examinations. Presence of any *Borrelia* in the blood is, however, confirmatory of the clinical syndrome of relapsing fever. Specimens taken during afebrile periods are likely to yield negative results.

RHEUMATIC FEVER (ACUTE RHEUMATIC FEVER, ACUTE RHEUMATISM)

1. *Etiologic agent.* Unknown. The upper respiratory tract infection which often precedes rheumatic fever is caused by Lancefield's Group A hemolytic streptococcus. Rheumatic lesions have not been found to contain an infecting organism; they resemble sensitization phenomena and are believed to be induced by a product of hemolytic streptococcal infection.

2. *Source of infection.* Unknown.

3. *Mode of transmission.* Unknown.

4. *Prevalence.* There is a close parallelism between the prevalence of rheumatic fever and the prevalence of recognized streptococcal respiratory tract infections. In the United States rheumatic fever is most prevalent in the Rocky Mountain region and in the New England and North and Central Atlantic states. The lowest incidence of rheumatic fever occurs in the south and southwest. The curve of seasonal incidence follows that of streptococcal infections, reaching its peak during the spring months and its low point during the summer and early autumn.

5. *Current laboratory services.* No specific laboratory aids to diagnosis known to Bureau of Laboratories.

RICKETTSIAL DISEASES (THE TYPHUS GROUP OF FEVERS)

A. Typhus

I. Epidemic or Classical Typhus (Louse-Borne)

1. *Etiologic agent.* *Rickettsia prowazeki,* var. *prowazeki.*

2. *Source of infection.* Infected persons.

3. *Mode of transmission.* The infectious agent is transmitted from man to man by lice *(Pediculus humanus)* which have fed upon infected persons. The rickettsias are inoculated by crushing the infected lice or scratching louse feces into the wound made by the bite or into other superficial skin abrasions. Dirty clothing contaminated with louse feces may be the source of infection transmitted through the air to the respiratory tract.

4. *Prevalence.* Widely distributed among people living under crowded and unhygienic conditions. Cases occur throughout the year with seasonal increase during the colder months.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Agglutination tests (Weil-Felix) on blood specimens.

   B. Complement-fixation tests for rickettsial diseases available at cooperating laboratories through the Bureau of Laboratories.

6. *Collection of specimens.*

   A. **For agglutination tests:** Collect aseptically about 5 ml. of venous blood 9 or 10 days after onset and place in sterile bottle in "TY" or "MI" outfit. Allow to clot firmly before sending to laboratory. Mark history blank plainly "For Weil-Felix test". Collect another specimen after the second week of illness.

   B. **For complement-fixation tests:** Use "MI" outfit. Collect aseptically 10-15 ml. of venous blood 9 or 10 days after onset and place in sterile bottle in outfit. Mark history blank plainly "For rickettsial diseases".

7. *Limitations of laboratory tests.* Weak or moderate results by the Weil-Felix test are of little diagnostic significance unless confirmed by a sharply rising titer during the third week of the disease. Negative results may be obtained as late as the 14th day of illness.

   Positive complement-fixation tests are confirmatory of clinical findings and usually indicate the type of infection present. The reaction is usually negative until the 9th or 10th day after onset.

II. Endemic or Murine Typhus (Flea-Borne)

1. *Etiologic agent. Rickettsia prowazeki,* var. *mooseri.*

2. *Source of infection.* Infected rodents, especially *Rattus rattus norvegicus.*

3. *Mode of transmission.* The agent is transmitted from rodent to man by a flea, commonly *Xenopsylla cheopis.*

4. *Prevalence.* Widely distributed in temperate, semitropical and tropical areas. Transmission to man occurs throughout the year, with seasonal increase during the warmer months.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Agglutination tests (Weil-Felix) on blood specimens.

   B Complement-fixation tests for rickettsial diseases available at cooperating laboratories through the Bureau of Laboratories.

6. *Collection of specimens.*

   A. **For agglutination tests**: Collect aseptically about 5 ml. of venous blood 9 or 10 days after onset and place in sterile bottle in "TY" or "MI" outfit. Allow to clot firmly before sending to laboratory. Mark history blank plainly "For Weil-Felix test". Collect another specimen after the second week of illness.

   B. **For complement-fixation tests**: Use "MI" outfit. Collect aseptically 10-15 ml. of venous blood 9 or 10 days after onset and place in sterile bottle in outfit. Mark history blank plainly "For rickettsial diseases".

7. *Limitations of laboratory tests.* Weak or moderate results by the Weil-Felix test are of little diagnostic significance unless confirmed by a sharply rising titer during the third week of the disease. Negative results may be obtained as late as the 14th day of illness.

   Positive complement-fixation tests are confirmatory of clinical findings and usually indicate the type of infection present. The reaction is usually negative until the 9th or 10th day after onset.

B. Rocky Mountain Spotted Fever (Tick-Borne)

1. *Etiologic agent. Rickettsia rickettsi.*

2. *Source of infection.* Infected ticks. In the eastern and southern United States the common vector is the dog tick, *Dermacentor variabilis;* in the northwestern United States, it is the wood tick, *Dermacentor andersoni*; in the southwestern United States it may occasionally be the lone star tick, *Amblyomma americanum.* In Brazil *Amblyomma cajennense* is the common vector. The rabbit tick *(Haemaphysalis leporis palustris)* has been found naturally infected, but this species does not bite man. The infection is passed from generation to generation in ticks and is probably maintained by infected and non-infected larvae feeding upon susceptible wild rodents.

3. *Mode of transmission.* Bite of tick or contact with tick material such as its blood or feces on the unbroken skin.

4. *Prevalence.* Known to occur throughout North America, and in several areas of South America. The season of occurrence is predominantly in the spring and early summer, corresponding to the time of appearance of adult ticks.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Agglutination tests (Weil-Felix) on blood specimens.

   B. Complement-fixation tests for rickettsial diseases available at cooperating laboratories through the Bureau of Laboratories.

6. *Collection of specimens.*

   A. **For agglutination tests:** Collect aseptically about 5 ml. of venous blood 9 or 10 days after onset and place in sterile bottle in "TY" or "MI" outfit. Allow to clot firmly before sending to laboratory. Mark history blank plainly "For Weil-Felix test". Collect another specimen after the second week of illness.

   B. **For complement-fixation tests:** Use "MI" outfit. Collect aseptically 10-15 ml. of venous blood 14-18 days after onset and place in sterile bottle in outfit. Mark history blank plainly "For rickettsial diseases".

7. *Limitations of laboratory tests.* Weak or moderate results by the Weil-Felix test are of little diagnostic significance unless confirmed by a sharply rising titer during the third week of the disease. Negative results may be obtained as late as the 14th day of illness.

   Positive complement-fixation tests are confirmatory of clinical findings and usually indicate the type of infection present. The reaction is usually negative until the 14th day after onset.

C. Tsutsugamushi Disease or "Scrub Typhus" (Mite-Borne)

1. *Etiologic agent. Rickettsia orientalis.*

2. *Source of infection.* Infected larval mites of *Trombicula akamushi* and related species varying with locality. The nymph and adult are free-living. The infection is passed from generation to generation and maintained by feeding upon susceptible wild rodents, particularly mice and rats of different species, varying with locality.

3. *Mode of transmission.* By the bite of infected mites.

4. *Prevalence.* Limited localities in countries of southeastern Asia, particularly India, Burma, Federated Malay States and French Indo-China; in the island archipelagoes of the west and south Pacific, Japan,

Formosa, Sumatra, Java, New Guinea, and in North Queensland, Australia. Near the Equator transmission may occur throughout the year; in Japan it is limited to the summer months.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Agglutination tests (Weil-Felix) on blood specimens.

   B. Complement-fixation tests for rickettsial diseases available at cooperating laboratories through the Bureau of Laboratories.

6. *Collection of specimens.*

   A. **For agglutination tests**: Collect aseptically about 5 ml. of venous blood 9 or 10 days after onset and place in sterile bottle in "TY" or "MI" outfit. Allow to clot firmly before sending to laboratory. Mark history blank plainly "For OXK agglutination". Collect another specimen after the second week of illness.

   B. **For complement-fixation tests**: Use "MI" outfit. Collect aseptically 10-15 ml. of venous blood 14-18 days after onset and place in sterile bottle in outfit. Mark history blank plainly "For rickettsial diseases".

7. *Limitations of laboratory tests.* Weak or moderate results by the Weil-Felix test are of little diagnostic significance unless confirmed by a sharply rising titer during the third week of the disease. Negative results may be obtained as late as the 14th day of illness.

   Positive complement-fixation tests are confirmatory of clinical findings and usually indicate the type of infection present. The reaction is usually negative until the 9th or 10th day after onset. In this disease complement-fixation tests serve the purpose of aiding in the elimination of typhus and Rocky Mountain spotted fevers.

D. Other Rickettsial Diseases

There are other diseases either proved to be caused or probably caused by one of the rickettsial agents. Q fever is caused by *Rickettsia burneti (Rickettsia diaporica)* which has been isolated from several species of ticks in Australia and the United States. Infections with this organism have been described as occurring in abattoir workers in Australia, and a laboratory outbreak characterized by atypical pneumonia was described in the United States.

Boutonneuse fever, a disease caused by *Rickettsia conori* and related to Rocky Mountain spotted fever, has extensive distribution in Rumania, Portugal, and the countries bordering the Mediterranean. It is also called Marseilles fever, *fièvre exanthematique, fièvre escarronodulaire.* Kenya typhus and South African tick fever may be closely related to boutonneuse fever. The dog apparently is an important reservoir.

The exact position of several of the so-called rickettsial diseases, such as "Indian tick-typhus", is not clear. Others such as "Trench fever" are not actually known to be rickettsial in origin.

Laboratory examinations may possibly be arranged through Bureau of Laboratories but can be performed only in laboratories especially equipped for the study of these infections. Contact Bureau of Laboratories before submitting any specimen. Special instructions will be given if arrangements for laboratory examinations can be made. In occasional instances laboratory examinations of diagnostic value may be possible.

RINGWORM (DERMATOPHYTOSIS)

A. Ringworm of the Scalp (Tinea Capitis)

1. *Etiologic agent.* *Microsporon audouini, M. canis (lanosum* or *felineum)* and other species of fungi cause sporadic tinea capitis.

2. *Source of infection.* Lesions on scalps of infected persons; articles of clothing, especially hats and caps containing the fungus or its spores, or infected hairs or scales shed by individuals. In the case of infection with *M. canis* or other animal fungi, contact with lesions or hair shed by young cats or dogs affected with ringworm.

3. *Mode of transmission.* Directly from person to person by contact with lesions of infected persons (or in the case of animal fungi, with infected animals). Possibly indirectly by articles of wearing apparel or by surfaces contaminated with scales or hairs from lesions. Transmission occurs in the home and in schools, especially during games in which personal contact is close.

4. *Prevalence.* Widespread, especially in school and institution outbreaks.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic and cultural examinations of hairs and skin scales from affected areas.

6. *Collection of specimens.* Collect hairs, preferably hair stumps, or scales from skin in a small clean envelope or on a clean, white sheet of paper. Seal envelope or fold paper to enclose material and place in suitable container such as "MI" outfit. Mark history blank plainly "For fungi".

7. *Limitations of laboratory tests.* Cultural identification of fungi observed microscopically is desirable.

B. Ringworm of the Body (including groin and feet)

1. *Etiologic agent.* Several species of fungi pathogenic to the skin.

2. *Source of infection.* Lesions on bodies of infected persons, articles of clothing carrying the fungus or its spores, or infected hairs or scales.

3. *Mode of transmission.* Directly by skin-to-skin contact with lesions of infected persons; possibly indirectly by articles of wearing apparel or towels or surfaces contaminated with scales or hair from such lesions.

4. *Prevalence.* Widespread, varying with conditions of crowding and during periods of warm weather.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic and cultural examinations of skin scales from the edge of lesions and skin scales from affected areas.

6. *Collection of specimens.* Collect skin scales from the edge of lesions or scales from skin in a small clean envelope or on a clean, white sheet of paper. Seal envelope or fold paper to enclose material and place in suitable container such as "MI" outfit. Mark history blank plainly "For fungi".

7. *Limitations of laboratory tests.* Cultural identification of fungi observed microscopically is desirable.

SANDFLY (PHLEBOTOMUS OR PAPPATACI) FEVER

1. *Etiologic agent.* A specific filterable virus.

2. *Source of infection.* The blood of an infected person.

3. *Mode of transmission.* The vector is a small, hairy, blood-sucking midge, *Phlebotomus papatasii,* which does most of its biting at night. It is possible that other species of *Phlebotomus* may also carry the virus.

4. *Prevalence.* Occurs only in those parts of Europe, Africa and Asia where the vector exists. In this respect it is a disease of subtropical and tropical areas where there are long periods of hot, dry weather, but in general it is to be found in a belt extending around the Mediterranean Sea eastward into Burma and China. It is seasonal, occurring between the months of April and October, and particularly prone to appear as a disease in troops which come from non-endemic areas and are quartered in the endemic zone during the spring and summer.

5. *Current laboratory services.* No laboratory aid to diagnosis is known to the Bureau of Laboratories.

SCABIES (THE ITCH)

1. *Etiologic agent.* *Sarcoptes scabiei,* the itch mite.

2. *Source of infestation.* Persons harboring the itch mite on their skin in burrows, particularly between the fingers.

3. *Mode of transmission.* Direct contact with infested person and indirectly by use of underclothing, gloves, bedding, etc. of such persons.

4. *Prevalence.* Widespread and occurring sporadically and in epidemics, under conditions of crowding, body uncleanliness, neglect, and lack of soap and water.

5. *Current laboratory services.* Identification of mites or of eggs, larvae or nymphs expressed from burrows in skin can be made through Bureau of Laboratories.

6. *Collection of specimens.* Place material in any convenient small vial containing 10% formalin and stopper tightly before submission to the laboratory.

7. *Limitations of laboratory tests.* Laboratory examination is seldom needed to establish or confirm a diagnosis of scabies.

SCHISTOSOMIASIS

1. *Etiologic agent.* Three species of schistosomes mature in man, *Schistosoma mansoni* in Central America, the West Indies, northern South America and Africa, *S. haematobium* in Africa, and *S. japonica* in the Orient. The ova of these three flukes are spined and are deposited in the abdominal venules from which they work their way to the mucosa of the bowel or bladder. None of these flukes is indigenous to the continental United States but they are found in Puerto Rico and the Philippines. The larvae of some other schistosomes found in the United States may cause "swimmer's itch" by penetrating the human skin. These schistosomes do not infect man and the larvae die in the skin.

2. *Source of infection.* Waters containing the intermediary snail host, contaminated by human excrement containing the ova of the parasite.

3. *Mode of transmission.* Ova hatch in the water and enter the snail host. In the snail multiplication occurs and swimming larval forms called "cercariae" develop, which leave the snail and, upon contact with the human skin, penetrate it to gain access to the blood stream.

4. *Prevalence.* No indigenous cases in the continental United States. Occurs in areas of the West Indies and northern South America; common in the Orient and Africa.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic examinations of feces and urine for identification of ova.

6. *Collection of specimens.* Collect portion of stool and urine specimens separately, placing them in bottles supplied in "PD" outfits. Mark history blank plainly "For schistosomes".

7. *Limitations of laboratory tests.* Identification of the ova is necessary to differentiate between species of *Schistosoma.* The ova of one species *(Schistosoma haematobium)* are usually found in the urine only.

SMALLPOX (VARIOLA)

1. *Etiologic agent.* A specific filterable virus.

2. *Source of infection.* Lesions of the mucous membranes and skin of infected persons.

3. *Mode of transmission.* By contact with persons sick with the disease; this contact need not be intimate, but aerial transmission except for short distances appears to be unlikely. By articles or persons freshly contaminated by discharges of the sick.

4. *Prevalence.* Distribution in sporadic or epidemic form; varies widely according to the immunity status of the population of an area and its exposure to infection from without. Occurrence is most frequent in the winter and least in summer months. There is no regional or climatic limitation to its prevalence except as population groups are more or less well protected by vaccination or by non-exposure to the disease.

5. *Current laboratory services.* None available at present at Bureau of Laboratories. (Culture of the virus is practicable only in specially equipped laboratories.)

STREPTOCOCCAL INFECTION — RESPIRATORY

A. Scarlet fever

B. Streptococcal Sore Throat, Streptococcal Nasopharyngitis, Streptococcal Tonsillitis, "Septic Sore Throat"

1. *Etiologic agent:*

   A and B: Hemolytic streptococci of Lancefield's Group A. Some strains, particularly in those which elaborate erythrogenic toxin (rash-producing factor) in high titer, produce either scarlet fever or the disease classified under B, depending upon the skin reactivity of the patient as determined by the Dick test. Other strains appear generally incapable of causing scarlet fever and produce only the other streptococcal infections, regardless of the Dick reaction of the patient.

2. *Source of infection:*

   A and B: Discharges from the nose, throat, or purulent complications of acutely ill or convalescent patients or carriers, or objects contaminated with such discharges.

3. *Mode of transmission:*

   A and B: Direct transmission can occur by contact with infected individuals during the incubation stage, during the acute infection, or during convalescence. Floor dust may be an important vehicle. Explosive outbreaks may follow the ingestion of contaminated milk or other foods.

4. *Prevalence:*

   A and B: Most common in the temperate zones, less common in the semi-tropical areas, and rare in tropical climates. Aside from food-borne epi-

demics, which may occur in any season, the highest incidence of scarlet fever in the United States occurs during the late winter and spring.

5. *Current laboratory services. Available at Bureau of Laboratories:*

A and B: Cultural examinations of swabbings from the mucosa of affected areas in the throat. Precipitin tests for grouping streptococci (Lancefield groups) are not made routinely but may be done when necessary to establish source of an outbreak. Typing of streptococci (Griffith types) is not done at present at Bureau of Laboratories.

6. *Collection of specimens:*

A and B: Use "HS" outfit. Rub sterile swab provided over reddened or otherwise affected areas in the throat. Remove stopper from tube containing agar and plunge swab into jelly and leave it there. Replace stopper and return to laboratory at once.

7. *Limitations of laboratory tests:*

A and B: A single negative culture should not be considered conclusive if there are clinical symptoms especially when more than 12 hours have elapsed between taking the culture and its receipt at the laboratory. In all such cases another culture should be sent.

Positive results on throat or nose cultures are not conclusive evidence that the beta hemolytic streptococcus is the cause of the condition. These results must be considered simply as contributory to the clinical evidence. Positive results on cultures from suppurating lesions are more conclusive.

STREPTOCOCCAL INFECTION — OTHER THAN RESPIRATORY

A. Erysipelas

1. *Etiologic agent.* Hemolytic streptococci of Group A.

2. *Source of infection.* Infected material from human sources containing hemolytic streptococci. Discharges from sinuses, draining ears or mastoids may cause erysipelas in adjacent tissues.

3. *Mode of transmission.* Bacteria entering breaks in skin either directly from same individual or indirectly from exogenous sources.

4. *Prevalence.* Geographic and seasonal distribution similar to that of streptococcal respiratory tract infections.

5. *Current laboratory services. Available at Bureau of Laboratories:* Cultural examinations of exudate from lesions or of pus from complicating suppurations. Precipitin tests for grouping streptococci (Lancefield groups) are not made routinely but may be done when necessary to establish source of outbreaks. Typing of streptococci (Griffith types) is not done at present at Bureau of Laboratories.

6. *Collection of specimens.* Use "HS" outfit. Collect exudate or pus on sterile swab in outfit. Remove stopper from tube containing agar jelly and plunge swab into jelly and leave it there. Replace stopper and return to laboratory at once.

7. *Limitations of laboratory tests.* The presence of streptococci producing beta hemolysis upon blood agar is strong presumptive evidence of their causative role in suspected erysipelas.

B. Puerperal Infection (Puerperal Septicemia)

1. *Etiologic agent.* Usually hemolytic streptococci of Group A. Infection may also be caused by anaerobic streptococci or by a mixed bacterial flora.

2. *Source of infection.* Hands and instruments used in examinations of the genital tract just prior to or dur-

ing or following confinement; the nose and throat of the parturient woman or her attendants just prior to, during, or just after confinement; infectious processes and discharges of the genital tract prior to confinement.

3. *Mode of transmission.* Direct transfer to the tissues of the parturient canal by hands, instruments, dressings, by droplets discharged in speaking, sneezing or coughing from infected or carrier individuals brought into close relation to the patient during or after delivery. Indirectly by articles soiled by infectious discharges brought into contact with the genital tract of the patient.

4. *Prevalence.* The most common cause of preventable sickness and death related to childbearing. Epidemics occur particularly in institutions where aseptic technics are faulty.

5. *Current laboratory services. Available at Bureau of Laboratories:* Cultural examinations of vaginal discharges.

6. *Collection of specimens.* Use "HS" outfit. Collect vaginal discharges on sterile swab in outfit. Remove stopper from tube containing agar, plunge swab into jelly and leave it there. Replace stopper and return to laboratory at once.

7. *Limitations of laboratory tests.* The presence of streptococci producing beta hemolysis on blood agar is confirmatory of clinical evidence.

SYPHILIS

1. *Etiologic agent. Treponema pallidum (Spirochaeta pallida).*

2. *Source of infection.* Discharges from obvious or concealed lesions of the skin and mucous membranes, the semen, the blood of infected persons, and, rarely, articles freshly soiled with such discharges or blood in which *Treponema pallidum* is present.

3. *Mode of transmission.* By direct contact with infected persons or through the blood of such persons; chiefly by sexual intercourse, occasionally by kissing; by dental and other surgical or technical accidents, and rarely by indirect contact with articles soiled with discharges containing the organism; congenitally from syphilitic mother through the placenta.

4. *Prevalence.* Widespread throughout the world varying with race, social customs, sex and age. Most commonly acquired between the ages of 18 and 30. Differences in racial incidence are related to social rather than biological factors.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Dark-field examinations of material from lesions for *Treponema pallidum.*

   B. Serodiagnostic tests on blood specimens.

   C. Serodiagnostic, total protein and colloidal gold tests on spinal fluids.

6. *Collection of specimens.*

   A. **For dark-field**: Use "CF" outfit. Wash lesion with sterile physiological salt solution. Rub firmly with sterile gauze. Remove all blood with sterile gauze. Gently compress the tissues surrounding the chancre until sufficient serum exudes to permit filling the capillary tubes. A compress of 2 per cent novacaine applied for a few

minutes will aid in obtaining the deep exudate. Hold the capillary tube in a horizontal position, while the specimen is being collected. The fluid will enter the tube rapidly due to capillary attraction. Seal by pressing both ends of tube into the wax in the small vial. Replace tube in the container and mail. If any antiseptic or other local treatment has been administered, a salt solution compress should be applied and the patient instructed to return on successive days. In case an antiseptic has been used on the lesion and the regional lymph glands are enlarged, fluid from them also may be submitted. In a bilateral adenitis there may be an associated chancroidal infection and one or more of the glands may be fluctuating, inflammatory and tender. The indurated, shotty, nontender glands should be chosen.

Sterilize skin over selected lymph node. Draw about 0.5 ml. of sterilized salt solution into a 1- or 2- ml. sterile syringe using a 22- or 24-gauge needle. Immobilize the gland by grasping it so that the skin is drawn tightly over it. Thrust the needle into the gland, rotate it to break apart some of the tissue at its tip, and inject a few drops of the salt solution, taking care that the point of the needle enters the gland and not the surrounding tissues. Draw into the syringe a few drops of the fluid from the glandular tissue. The aspirated fluid (which should contain very little blood) may then be deposited from the syringe onto any glass surface, such as that of a microscope slide or the clean side of a flat bottle, and collected in capillary tubes in the same manner as described for fluid from a chancre.

B. **Blood specimens**: Use "SY" outfit. Collect about 5 ml. of blood in the dry, sterile, chemically clean jar provided in the container. If a syringe is used it should be dry. Never use a hot syringe. Never use a syringe or a needle containing any trace of water, alcohol or oil. If syringe and needle cannot be thoroughly dried, rinse with sterile physiological salt solution. Avoid air bubbles in blood when drawing specimen; do not force blood violently from syringe into specimen vial. Replace cover or cork promptly after putting blood in vial.

Always allow clot to form firmly in the vial before blood is agitated in any way. Do not expose blood to heat such as placing near a radiator or sterilizer or placing in direct sunlight: do not expose to freezing temperatures. Mail or otherwise send specimens promptly to the laboratory; avoid when possible shipping specimens over a week-end or a holiday. When specimens cannot be mailed or sent within an hour or two, place in a refrigerator but not where it will freeze.

C. **Spinal fluid**: Use "SF" outfit. Collect about 10 ml. of spinal fluid. Discard enough of first flow of fluid to eliminate blood present in needle which may have passed through small blood vessel. Let fluid flow directly into sterile bottle supplied with this outfit; replace cap tightly. Keep fluid in refrigerator until shipment to laboratory; do not remove blood or other material by centrifuging. Mark history blank plainly to indicate type of examination requested.

7. *Limitations of laboratory tests.*

A. **Dark-field examinations**: Positive results on specimens from the mouth should be viewed with caution since oral spirochetes have a morphology similar to *Treponema pallidum*. The report of a trained observer will always take this into consideration.

Negative results on specimens from infected individuals may result when antiseptics have been applied locally.

B. **Blood tests**: No single test for syphilis will pick up all cases which may prove positive by another standard test. Non-specific or false reactions infrequently occur in healthy individuals but may occur in febrile conditions or in malaria and a few other diseases relatively rare in Connecticut. The physician may, however, frequently encounter the asymptomatic case of lues. For these reasons, the following suggestions

are made for evaluating laboratory results obtained in Bureau of Laboratories where two separate tests are made on reacting specimens:

1. **Both tests positive.** Repeat the test if careful clinical examination and negative history of exposure are at variance with laboratory results.

2. **One test positive, one doubtful or negative.** Always take second specimen for repeat test. A repeated positive test, supported or not by another type of test, constitutes a positive serology but the final diagnosis can be made by the physician only with full consideration of clinical findings.

3. **One test doubtful, one negative (or both doubtful).** Always take second specimen for repeat test. Even consistently repeated findings of this nature are not always indicative of infection but do require careful clinical investigation. Clinical findings should outweigh laboratory findings.

4. **Repeating tests.** When submitting repeat specimens on cases without clinical evidence of syphilis, mark history blank plainly "For special studies".

5. **Negative test.** Repeat if suspected primary case. Reactions may not develop until 5-8 weeks after primary lesion appears.

C. **Tests on spinal fluid**: A positive serodiagnostic test for syphilis is considered indicative of central nervous system involvement since false reactions occur rarely or not at all with spinal fluid in a competent laboratory performing a standard test. Cell counts (preferably made locally), total protein determinations and colloidal gold tests are helpful in determining extent and type of involvement.

TETANUS

1. *Etiologic agent.* Tetanus bacillus, *Clostridium tetani.*
2. *Source of infection.* Soil, street dust, and animal feces.
3. *Mode of transmission.* Wound infection.
4. *Prevalence.* World-wide distribution, following wound infection. Most frequent in North America among young males and in summer. Prevalent especially following wounds contaminated with manured soil. Tetanus in the new born occurs where there is neglect of surgical asepsis and ordinary cleanliness in the care of the umbilical cord and its dressings in the first two weeks of life. The condition is a serious factor in infant mortality where midwives are ignorant or incompetent.
5. *Current laboratory services. Available at Bureau of Laboratories:* Cultural examinations and animal inoculation tests of curettings from wounds.
6. *Collection of specimens.* Place tissue fragments from curetted wound in sterile bottle in "MI" outfit. Mark history blank plainly "For tetanus".
7. *Limitations of laboratory tests.* Animal inoculation tests are more reliable than cultures; a negative culture is not conclusive.

TRACHOMA

1. *Etiologic agent.* A filterable virus.

2. *Source of infection.* Secretions and purulent discharges from the conjunctivae and adnexed mucous membranes of the infected persons.

3. *Mode of transmission.* By direct contact with infected persons and indirectly by contact with articles freshly soiled with the infective discharges of such persons.

4. *Prevalence.* World-wide, appearing in parts of all continents and most islands of the Pacific. Cases most common among children but may occur and persist at any age.

5. *Current laboratory services.* No reliable laboratory aids to diagnosis known to Bureau of Laboratories.

TREMATODE INFECTIONS, MISCELLANEOUS

1. *Etiologic agent.* Among others are *Fasciola hepatica* (sheep liver fluke), *Fasciolopsis buski* (giant intestinal fluke), *Clonorchis sinensis* (Chinese liver fluke), *Paragonimus westermani* (Oriental lung fluke), *Echinostoma ilocanum* (Garrisons fluke), *Heterophyes heterophyes* (minute fluke), *Metagonimus yokogawai* (Yokogawa's fluke).

2. *Source of infection.* Infected humans or other animals that may serve as a primary host.

3. *Mode of transmission.* All of the flukes have a complex life cycle and infection of the human takes place by ingestion of the infective form of the cercaria in water, on aquatic plants, in raw vegetables, fish, etc.

4. *Prevalence.* For the most part limited to oriental regions. *Fasciola hepatica* is present in Europe and in the western hemisphere.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic examinations of feces for ova; similarly, examinations of sputum for ova of lung fluke *(Paragonimus).*

6. *Collection of specimens.* Use "PD" outfit. Place material in bottle furnished. Mark history blank plainly "For trematodes".

7. *Limitations of laboratory tests.* Identification of species by study of the ova is possible by an observer with special training.

TRICHINOSIS

1. *Etiologic agent. Trichinella spiralis.*

2. *Source of infection.* Uncooked or insufficiently cooked pork or pork products.

3. *Mode of transmission.* Only through consumption of meat containing viable larvae.

4. *Prevalence.* World-wide. The parasite is particularly widespread in the United States, about 1 in every 6 necropsies showing infection. Clinical cases probably occur more frequently than is indicated by morbidity reports and the disease is probably often confused with other illnesses. No selection by age, sex, race, region, season, or climate except as these affect the custom of eating the insufficiently cooked flesh of infected hogs.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Examination of blood smears for eosinophilia.

   B. Precipitin tests on blood serum, preferably during fourth week of disease, available through cooperating laboratories.

   C. Examination of laked blood for larvae of *Trichinella spiralis.*

   D. Examination of biopsy tissue (usually from deltoid or pectarol muscle) obtained after 10th day of illness for encysted trichina.

   E. Examination of suspected meat or meat products for larvae of *Trichinella spiralis.*

6. *Collection of specimens.*

   A. Collect thin blood smears. Use "MA" outfit. Follow directions given under "MALARIA" for thin smears only. Mark history blank plainly "For trichinosis".

   B. **Blood**: Use "MI" outfit. Collect aseptically about 5 ml. of venous blood and place in sterile bottle in outfit. Allow to clot firmly before sending to laboratory. Mark history blank plainly "For trichinosis".

   C. In severe cases during the time of high fever (second or third week) collect 5 ml. of venous

blood and add immediately to 15 ml. of 3% acetic acid. A suitable outfit for collection of the blood will be furnished upon request to the Bureau of Laboratories. Mark history blank plainly "For trichina".

D. In cases which develop severe pain, stiffness and disability (usually about the 10th day of illness), the physician may in rare instances wish to excise a bit of muscle from the deltoid or pectoral muscle near its insertion for examination for encysted trichinae. Place excised tissue in sterile bottle in "MI" outfit. Mark history blank plainly "Biopsy material for trichina".

E. **Meat**: Submit a representative sample of the suspected meat in any convenient container. Mark accompanying request plainly "For trichina".

7. *Limitations of laboratory tests:*

A. **Eosinophilia**: Although not specific, marked eosinophilia lends support to clinical evidence of this disease. It occurs early enough to be a distinct diagnostic aid.

B. **Precipitin test**: Since this reaction does not usually occur until after the third week of the disease, it is of limited value as a diagnostic aid.

C. **Trichinae in blood**: It is possible to demonstrate the trichinae in laked blood ordinarily only in more severe cases. A negative result is not conclusive.

D. **Trichinae in biopsy tissue**: This test of distinct diagnostic value but is not recommended except when absolutely necessary to establish a diagnosis.

E. **Trichinae in meat**: It is sometimes very difficult to establish the presence of trichinae in suspected meat, particularly ground pork products, and the examination must be carried out painstakingly. A negative result is not entirely conclusive.

Note: During the early acute gastro-enteritis adult worms may occasionally be found in feces or in vomitus. However, although present in the intestinal contents at autopsy of fatal cases, the worms are usually marred beyond recognition in body discharges during life.

TRICHURIS INFECTION (WHIPWORMS)

1. *Etiologic agent.* *Trichuris trichiura,* the whipworm.
2. *Source of infection.* Infected humans.
3. *Mode of transmission.* Ingestion with food or otherwise of ova from soil contaminated by excretions. Flies may be an intermediate factor in contaminating food.
4. *Prevalence.* Widely distributed in warm moist regions where general sanitation is poor.
5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic examinations of feces for ova.
6. *Collection of specimens.* Use "PD" outfit. With a tongue depressor or similar implement take specimens of feces from different parts of the stool shortly after it is passed and place in bottle supplied with outfit.
7. *Limitations of laboratory tests.* *Trichuris trichiura* infection is usually asymptomatic unless very heavy. The worm may be present together with other parasites.

TRYPANOSOMIASIS, AMERICAN* (CHAGAS' DISEASE)

1. *Etiologic agent.* *Trypanosoma cruzi.*

2. *Source of infection.* Infected persons and a number of domestic and wild animals, such as dogs, cats, opossums, and armadillos.

3. *Mode of transmission.* Fecal material of infected insect vectors, various blood sucking species of Reduviidae (cone-nosed bugs), especially the genus *Triatoma*, which frequently attack man. Contamination with infected fecal material from the bug, of the conjunctivae, mucous membranes, abrasions, or wounds in the skin made by the bite of the insect. It is probably not transmitted by the actual act of biting.

4. *Prevalence.* The disease has a wide geographic distribution in Central and South America. Cases have been found in Southern Mexico. No human case has been reported as yet in the United States but several species of the insect genus *Triatoma* have been shown to be carriers of *Trypanosoma cruzi* in Texas, New Mexico, Arizona, and California.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic examinations of thick blood films and citrated blood, and animal inoculation tests on fluid blood.

6. *Collection of specimens.*

   A. Collect thick blood films as for malaria (See "MALARIA"). Use "MA" outfit. Mark history blank plainly "For trypanosomes".

   B. Collect citrated blood for microscopic examination and for animal inoculation as follows: Withdraw 9 ml. venous blood and add it immediately to tube or bottle containing 1 ml. of 6% sodium citrate solution. Mix well but gently. A special outfit for this purpose will be furnished by the Bureau of Laboratories upon request. Mark history blank plainly "For trypanosomes".

7. *Limitations of laboratory tests.* These examinations are reliable diagnostic aids in the hands of a specially trained worker.

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* The laboratory services described apply equally well to African trypanosomiasis.

TUBERCULOSIS, PULMONARY

1. *Etiologic agent.* Tubercle bacillus (human), *Mycobacterium tuberculosis (hominis);* bovine type has been found in occasional cases in some areas (outside the continental United States) where milk is not pasteurized and infection of cattle is prevalent; avian type doubtful for human infections.

2. *Source of infection.* Persons with "open" pulmonary tuberculosis; rarely tuberculosis cattle.

3. *Mode of transmission.* Usually through the discharges of the respiratory tract, by direct or indirect contact with infected persons, by means of coughing, sneezing, or other droplet infection, by kissing, by the use of contaminated eating and drinking utensils, and possibly by contaminated flies and dust. Infection rarely occurs from casual contact, but usually results from the continued type of exposure characteristic of family relationships.

4. *Prevalence.* Among the most common communicable diseases of man, with less variation in prevalence of infection according to race than in mortality. In most occidental nations its incidence and mortality are declining. Age at which first infection occurs varies; children exposed in the household and in cities are infected earlier than rural children and those not so exposed, who may escape infection until adolescence or adult age. Mortality high among infants, among adult males up to old age, and among adolescent and young adult females. Leading cause of death at ages 15 to 34. Aboriginal races when first exposed develop the disease in a rapidly fatal form. The disease occurs at times in epidemics.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic and cultural examinations of sputum for *Mycobacterium tuberculosis.*

6. *Collection of specimens.* Sputum should be collected in the clean, wide-mouthed jar fitted with a tight cover that is supplied in the container. Purulent, cheesy or mucopurulent sputum, preferably that coughed up from the lungs early in the morning, is much more likely to contain tubercle bacilli than just mouth saliva. The presence of saliva, mucous, blood or stomach contents in the specimen is undesirable for reliable examination.

7. *Limitations of laboratory tests.* Failure to find *Mycobacterium tuberculosis* microscopically, even with concentration methods, does not exclude pulmonary tuberculosis. Positive microscopic findings occasionally may be due to the presence of acid-fast bacilli other than those of tuberculosis. Cultural confirmation of results is desirable in either case.

Negative cultural findings are of no value if preservative has been added to the original specimen. In the absence of preservative, negative cultural findings on specimens which are positive microscopically are unusual and a repeat specimen should be submitted; when acid-fast organisms other than tubercle bacilli are isolated from such specimens, a special report will be sent. A negative microscopical or cultural result does not exclude the possibility of tuberculosis infection.

Positive cultural findings on specimens which were negative microscopically do not constitute a conflicting report since the cultural method is a more sensitive means of detecting small numbers of tubercle bacilli than is the microscopic examination although it requires a much longer time for completion. Animal inoculation tests of cultures isolated are often desirable to demonstrate that the culture of acid-fast organisms isolated has been found to possess the virulence characteristic of *Mycobacterium tuberculosis.*

TUBERCULOSIS OTHER THAN PULMONARY

1. *Etiologic agent.* Tubercle bacillus (human and bovine), *Mycobacterium tuberculosis (hominis et bovis).*

2. *Source of infection.* Persons with "open" pulmonary tuberculosis, less frequently tuberculous cattle.

3. *Mode of transmission.* By direct contact with infected persons, by contaminated food, and possibly by contact with articles freshly soiled with the discharges of infected persons.

4. *Prevalence.* Much less common than the pulmonary form and more rapidly falling in incidence, representing in the United States less than 10 per cent of total cases and deaths from the disease. Especially common in infants and young children where intimately exposed to parental infection and to bovine infection through unpasteurized milk from tuberculous cattle.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic examinations and animal inoculation tests on pus, exudates, and body fluids for presence of *Mycobacterium tuberculosis.*

6. *Collection of specimen.* Collect material aseptically and place in sterile bottle in "MI" outfit. Mark history blank plainly "For animal inoculation for tuberculosis". Use "SF" outfit for collection of spinal fluid in cases of suspected tuberculous meningitis.

7. *Limitations of laboratory tests.* Positive microscopic results are suggestive of infection: positive animal inoculation tests are conclusive. Negative microscopic findings may be of little value but negative animal inoculations are reliable in most instances when specimens have been properly collected.

TULAREMIA

1. *Etiologic agent.* *Pasteurella tularensis (Bacterium tularense).*

2. *Source of infection.* Wild rabbits and hares, horse fly *(Chrysops discalis)*, wood tick *(Dermacentor andersoni* and *Dermacentor variabilis)*, woodchuck, coyote, muskrat, opossum, tree squirrel, quail, skunk, water rat of Europe *(Arvicola amphibus)*, cat, deer, dog, fox, hog, sage hen, and bull snake.

3. *Mode of transmission.* By bites of infected flies and ticks and by inoculation through handling infected animals, as in skinning, dressing, or performing necropsies on infected animals, or by fluids from infected flies, ticks, rabbits, and woodchucks. Ingestion of insufficiently cooked rabbit meat. Rare cases occur from bites of coyotes, skunks, hogs, cats, and dogs, where the mouth of the animal was presumably contaminated from eating infected rabbits. Drinking contaminated water. Infections acquired in the laboratory are not infrequent.

4. *Prevalence.* The disease occurs throughout North America, in many parts of continental Europe and in Japan. In the United States it occurs in every month of the year, but especially during the rabbit hunting season. The case fatality is about 5 per cent.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Agglutination tests on blood specimens.

   B. Bacteriological examinations of material from lesions are not ordinarily recommended but can

be arranged for through cooperating laboratories under unusual conditions.

C. Bacteriological and pathological examination of animals or organs from animals suspected of transmitting tularemia.

6. *Collection of specimens.* After the 10th day of illness, collect aseptically 5-10 ml. of venous blood in the sterile bottle in "TY" outfit; mark history blank plainly "For tularemia".

Do not collect any other type of specimen until you have contacted Bureau of Laboratories for special instructions.

7. *Limitations of laboratory tests.*

A. **Agglutination tests**: The agglutination test for tularemia does not become positive until the 10th day of illness and reaches its maximum reaction 14-18 days after onset. The test remains positive indefinitely following infection; hence, previous history is important. False reactions are known to occur only in brucellosis.

B. **Bacteriological studies**: Demonstration of *Pasteurella tularensis* in material from lesions is a difficult laboratory procedure involving extreme danger to laboratory workers. It is not a recommended procedure.

C. **Animal sources**: A trained animal pathologist can recognize the lesions of tularemia in rodents by gross study at autopsy. Bacteriological studies on animals and animal organs are subject to the same dangers discussed under "B" above.

TYPHOID FEVER

1. *Etiologic agent.* Typhoid bacillus, *Eberthella typhi.*

2. *Source of infection.* Bowel discharges and urine of infected individuals and carriers. About 2 to 5 per cent of patients become permanent carriers. Family contacts may be transient carriers.

3. *Mode of transmission.* Conveyance of typhoid bacilli by direct or indirect contact with patient or carrier. Among indirect means of transmission are contaminated food, water, milk, and shellfish, and, under some conditions, flies.

4. *Prevalence.* Widespread throughout the world. Formerly endemic and epidemic in most large cities of North America and in many rural areas. Still endemic in some rural areas of the United States, but commonly now occurring as sporadic cases and as small contact and carrier epidemics. Steadily falling in incidence, particularly in urban areas supplied with safe water and pasteurized milk, and where human excreta are disposed of without contaminating water supplies, food, milk, or surface of the soil.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Blood cultures (1-7 days' duration of disease).

   B. Agglutination tests (after 10th day of illness). Cultures are made from the clots after the serum has been removed.

   C. Feces, urine and bile cultures for isolation and typing of organism.

   D. Tests to detect Vi agglutinins in blood of suspected carriers.

   E. Bacteriophage typing of typhoid cultures for epidemiological purposes made routinely on all cultures isolated at Bureau of Laboratories.

6. *Collection of specimens.*

   A. **Blood cultures**: Use "BC" outfit. Collect aseptically venous blood for culture in sterile syringe of 5-10 ml. capacity. Preferably replace needle used with another sterile unused one. Plunge needle through thin rubber top of stopper (previously swabbed with alcohol) and expel con-

tents of syringe into medium in bottle. Rush to laboratory.

B. **Agglutination tests and clot cultures**: Collect 5-10 ml. venous blood aseptically in sterile bottle in "TY" outfit. Allow to clot firmly before sending to laboratory.

C. **Feces**: Use "FE" outfit. Using the sterile swab in the outfit, transfer a portion of the stool about the size of a sphere 1/2 inch in diameter to the specimen jar and emulsify it in the glycerol solution. If stools are liquid, transfer to the specimen jar with a clean spoon or other implement a quantity approximately 2 ml. in volume. Do not use spoon or other implement for more than one sampling unless carefully sterilized in the interim. Care should be taken that the stool is passed into a vessel that has been thoroughly disinfected or sterilized.

Urine: Use "UR" outfit. Have patient void urine into previously sterilized vessel and place 10-15 ml. in sterile bottle provided in outfit.

Bile: Collect this material ordinarily only when carrier has been hospitalized for release from carrier state under the provisions of the Sanitary Code of Connecticut. Use duodenal tube and place 10-15 ml. of biliary drainage in sterile bottle in "FE" outfit. Be sure to check history blank to indicate specimen is bile.

D. **Blood for Vi agglutinins**: Follow directions under "B" above; mark history blank plainly "For Vi agglutinins".

E. **Bacteriophage typing**: Heads of local laboratories may submit cultures for this service.

7. *Limitations of laboratory tests.*

A. For early diagnosis a blood culture is the best diagnostic aid. It should be taken during the first five days of illness, after which the organisms usually disappear from the blood stream. Isolation of a serologically typed *Eberthella typhosa (Salmonella typhosa)* establishes the cause of illness.

B. Agglutinins do not appear in the blood until 10-14 days after onset. The titer reaches its height during the third week of illness. Agglu-

tinins produced as a result of previous infection or of prophylactic vaccination may persist for an indefinite period of time. Typhoid "O" agglutinins resulting from immunization do not ordinarily persist longer than 3-6 months after inoculation but typhoid "H" agglutinins may persist longer. Hence, a high typhoid "O" titer is of considerable significance when clinical findings are suggestive. Such findings must, however, be confirmed by positive blood or feces cultures.

Results of clot cultures have the same significance as those on blood cultures (See A. above). Negative results are, however, likely to occur after the first five days of illness.

C. Isolation of serologically typed *Eberthella typhosa (Salmonella typhosa)* from the feces, urine or bile may mean the patient is a temporary or chronic carrier or that the organism isolated is the cause of the illness. Hence, the significance of positive results must be considered in the light of clinical findings. For release of cases and of carriers, see the requirements of the Sanitary Code of Connecticut.

D. Absence of Vi agglutinins is not conclusive evidence of absence of carrier state. Presence of Vi agglutinins simply directs attention to possible carriers for cultural confirmation. The test is of most value for rapid screening purposes among large numbers of suspects.

E. It is well established that typhoid strains do not vary with respect to bacteriophage type in establishing relationship between cases and carriers.

UNDULANT FEVER (BRUCELLOSIS)

1. *Etiologic agent. Brucella melitensis (Alkaligenes melitensis, Micrococcus melitensis); Brucella abortus (Alkaligenes abortus); Brucella suis.*

2. *Source of infection.* The tissues, blood, milk, and urine of infected animals, especially goats, cattle, and swine. Laboratory infections take place readily.

3. *Mode of transmission.* By ingestion of milk from infected animals and by direct contact with infected animals or animal products.

4. *Prevalence.* Occurs more often in males than in females, and particularly in persons whose occupation brings them into direct contact with milk cows, hogs, or goats, and in persons using unpasteurized milk of cows or goats. Found in every one of the United States and in Canada, affecting persons of all races. Occurs most often in the months of May to October. Many cases of a mild type doubtless occur without record.

5. *Current laboratory services. Available at Bureau of Laboratories:*

   A. Agglutination tests on blood specimens.

   B. Blood cultures for *Brucella*; cultures routinely made on clots from blood specimens submitted for agglutination tests.

   C. Opsonocytophagic tests for brucellosis.

   D. Special studies on suspected sources of infection available through special arrangement.

6. *Collection of specimens.*

   A. **For agglutination tests and clot cultures**: Use "TY" outfit. After the 10th day of illness, collect aseptically 5-10 ml. of venous blood and place in sterile bottle in outfit.

   B. **Blood cultures**: Use "BC" outfit. During febrile episode, prepare the skin at the bend of the elbow with iodine and alcohol. Place a tourniquet on the arm. Have the patient extend the arm fully and open and close the fist a few times to distend the veins. With the sterile syringe and needle, remove not less than 5 ml. Have the pa-

tient open his fist, then remove the tourniquet, and withdraw the needle. Preferably replace used needle with another unused sterile one. To transfer blood to the culture outfit pass the needle of the syringe through the rubber diaphragm in the top of the screw cap (foil covering having been previously removed), and force the blood directly into the bottle. The enriched medium in the container will prevent clotting of blood and support growth of organisms that may be present. It is important that not less than 5 ml. or more than 10 ml. of blood be added. The outfit should be returned to the laboratory as soon as possible.

C. **Blood for opsonocytophagic test**: Have this test done before injecting skin test antigen. Obtain venous blood using aseptic precautions and immediately add exactly 5 ml. to the vial. Replace stopper and invert vial to mix thoroughly. Return to Laboratories within eight hours after collection. Use messenger if special delivery mail service or bus service will not permit specimen to arrive during working hours. Always state time of collection on history blank since specimens over eight hours old are entirely unreliable due to disintegration of leucocytes.

D. **Milk and similar materials**: Contact Bureau of Laboratories for special instructions before sending any specimens or samples.

7. *Limitations of laboratory tests.*

A. **Agglutination tests**: Strong agglutination is highly suggestive and confirmatory of suspected brucellosis. Weak reactions may have significance in many cases. Some infected persons never develop detectable agglutinins; hence, negative findings are not conclusive.

B. **Blood cultures and clot cultures:** Since *Brucella* are present in the blood stream in detectable numbers only during unpredictable "showers" and since this is most likely to occur during febrile episodes, negative results on a single specimen are not conclusive. A series of specimens is recommended. Positive findings are diagnostic.

C. **Opsonocytophagic tests**: Injection of skin test antigen before this test is done may produce confusing results. The results of the opsonocytophagic test must be considered with the skin test reaction before conclusions can be drawn. Individuals with negative skin tests have not been sensitized to *Brucella* and are probably susceptible to infection but not infected. Individuals with positive skin tests may be either infected or immune. Those who are infected may show no phagocytic activity of the blood or varying degrees up to 40% marked phagocytosis. Those who are definitely immune show 60% or more marked phagocytosis. Those who show between 40% and 60% marked phagocytosis may be classed as "questionably infected" and repeat specimens should be taken when necessary. Presumably, those classed as "questionably infected" are either developing immunity or are losing an immunity established previously. The status of individuals who have lost a considerable degree of immunity but have not been re-infected is not clear.

D. **Special tests on milk, etc**: Demonstration of agglutinins in milk serum or isolation of the organism from milk or similar products provides evidence of epidemiological value only.

VINCENT'S INFECTION (TRENCH MOUTH), FUSOSPIROCHETOSIS

1. *Etiologic agent.* Complex; fusospirochetal flora (fusiform bacteria and *Borrelia vincentii*) proliferates rapidly under pathologic conditions.

2. *Source of infection.* Discharges from the lesions of infected persons or from carriers.

3. *Mode of transmission.* Direct contact with infected persons or carriers and probably with articles freshly soiled by such persons.

4. *Prevalence.* Sporadic in general population. More common among persons of low nutrition and neglected hygiene. More common in children and young adults, and often occurs in outbreak form.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic examination of smears of exudate or pus from affected areas.

6. *Collection of specimens.* Use "VI" outfit. With sterile swab provided collect exudate or pus from affected areas and make smears on microscope slides provided. The use of a wire loop for the collection of material for examination is sometimes convenient.

7. *Limitations of laboratory tests.* The organisms usually associated with Vincent's infection are sometimes found on apparently healthy mucous membranes, particularly those of the mouth, in the absence of lesions. For that reason a positive result should be interpreted with caution, placing the most reliance upon the clinical picture. Since these organisms grow readily in diseased tissues, their presence is not proof of a primary etiological role.

VULVOVAGINITIS IN CHILDREN

1. *Etiologic agent.* A variety of organisms, including the *Neisseria gonorrhoeae.*

2. *Source of infection.* Discharges of infected persons.

3. *Mode of transmission.* By direct contact with infected persons and by contact with articles freshly soiled with the discharges of such persons. In children, usually spread by other than sexual contact.

4. *Prevalence.* Widespread; most common in families where there are overcrowding, neglect in personal cleanliness, and ignorance as to sanitary precautions. Epidemics are observed most frequently in child-caring institutions, day nurseries and schools.

5. *Current laboratory services. Available at Bureau of Laboratories:* Microscopic and cultural examinations of discharges for causative organism, particularly *Neisseria gonorrheae* (the gonococcus).

6. *Collection of specimens.*

   A. **Smears**: Use "GC" outfit. Collect vaginal discharges on swab provided and make thin smears on microscope slides. Allow smears to dry before shipment to laboratory.

   B. **Gonococcal cultures**: Use "NC" outfit. Collect vaginal discharges on small swab inserted in the cork of the outfit. Place in tube containing blood-dye mixture using cork as stopper. Rush to laboratory.

   C. **Cultures for other organisms**: Use "HS" outfit. Collect vaginal discharges on sterile swab provided, then plunge swab into agar jelly in tube furnished. Stopper and send to laboratory. Mark history blank plainly "For identification of organism".

7. *Limitations of laboratory tests.* Gram-negative intracellular diplococci observed microscopically can be confirmed as gonococci only by gonococcal cultures. Presence of other pathogenic bacteria is presumptive evidence of etiological significance.

YAWS (FRAMBESIA)

1. *Etiologic agent.* *Treponema pertenue.*

2. *Source of infection.* Discharges from skin lesions and mucous membranes.

3. *Mode of transmission.* Direct contact with lesions of patient and by nonbiting flies which convey the discharges of infected persons to others.

4. *Prevalence.* Very common in the tropics, especially in Africa, Polynesia, the Philippines, and some parts of the Western hemisphere. In the West Indies more prevalent in some villages than others. At present not known to be indigenous in continental North America. Especially prevalent in the Caribbean area: Jamaica, Haiti, Trinidad, Antigua, and other islands of the Leeward group, and in some coastal and valley settlements of Colombia.

5. *Current laboratory services.* See "SYPHILIS".

6. *Collection of specimens.* See "SYPHILIS".

7. *Limitations of laboratory tests.* There is no reliable way of distinguishing between the *Treponema pertenue* of yaws and the *Treponema pallidum* of syphilis or between the serological reactions evoked by both.

YELLOW FEVER

1. *Etiologic agent.* A specific filterable virus.

2. *Source of infection.* The blood of infected persons, monkeys, marmosets, and probably some other wild animals.

3. *Mode of transmission.* By the bite of infected *Aedes aegypti* mosquitoes and of a number of bush mosquitoes of which *Hemagogus* appears to be the most important in South America. (It is not yet certain that some other biting insect may not be capable of acting as the transmitter.)

4. *Prevalence.* Not known in the Pacific Basin. No case in North America or Puerto Rico for many years. Endemic among human beings and some animals of western and central Africa. Endemic in certain species of monkeys (and perhaps other jungle animals) of northern and central South America, and probably in eastern Panama. Occasional epidemics occur among human beings; the infection transmitted in towns by *Aedes aegypti* and in the bush by other mosquitoes; sporadic human cases, probably of jungle origin.

5. *Current laboratory services.* None available at present at Bureau of Laboratories. Special serological tests and other virus studies are done at a few virus laboratories throughout the country.

6. *Collection of specimens.* Contact Bureau of Laboratories to determine if studies at a cooperating laboratory can be arranged; instructions will be given at that time.

7. *Limitations of laboratory tests.* Lack of facilities constitutes the main limitation at present. Demonstration of increasing titer of neutralizing antibodies during course of disease is of diagnostic value.

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